1986

The use of Crab Meal as a Supplemental Food for Juvenile Hard Clams (Mercenaria mercenaria)

Patricia Lynn Duncan
College of William and Mary - Virginia Institute of Marine Science

THE USE OF CRAB MEAL AS A SUPPLEMENTAL FOOD FOR JUVENILE HARD CLAMS (*MERCENARIA MERCENARIA*)

A Thesis

Presented to

The Faculty of the School of Marine Science

The College of William and Mary in Virginia

In Partial Fulfillment

Of the Requirements for the Degree of

Master of Arts

---

by

Patricia L. Duncan

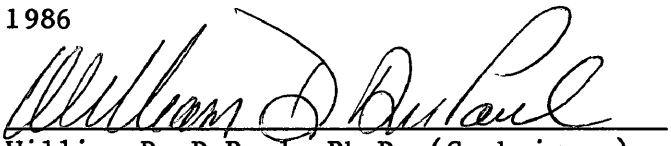
1986

This thesis is submitted in partial fulfillment of
the requirements for the degree of

Master of Arts

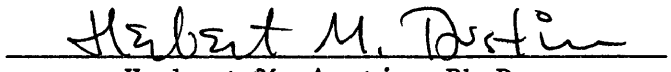
Patricia L. Duncan

Approved, August 1986

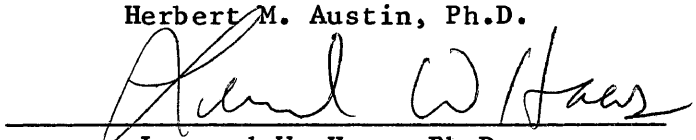


William D. DuPaul, Ph.D. (Cochairman)

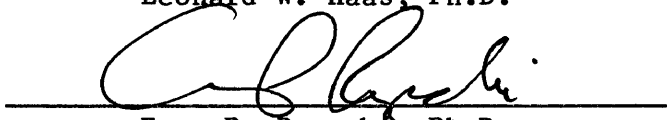
Michael Castagna, M.S. (Cochairman)



Herbert M. Austin, Ph.D.



Leonard W. Haas, Ph.D.



Evon P. Ruzecki, Ph.D.

TABLE OF CONTENTS

ACKNOWLEDGMENTS

I wish to express my appreciation to Dr. Bill DuPaul and Mike Castagna, cochairmen of my thesis committee, for guidance during the course of this study and for helpful criticism during the writing of this thesis. I especially want to thank Bill DuPaul for suggestion of this research topic. I would also like to thank my other committee members, Drs. Larry Haas, Herb Austin, and John Ruzecki for helpful suggestions throughout the study and for review of the manuscript. Additional thanks go to Larry Haas for instruction and help with epifluorescent microscopy. Special thanks go to Jim Moore of the Wachapreague Lab for help in the design and construction of the experimental apparatus. Thanks to Weston F. Conley, Jr. for supplying the crab meal. I also want to thank Dr. George Flick and Deborah Holloway of the Department of Food Science and Technology of Virginia Polytechnic Institute and S.U. for determining the proximate analysis and amino acid content of crab meal. I also wish to express grateful appreciation to the following VIMS personnel: Dr. Mary Gibbons for reviewing the manuscript; Cindy Fischler for help with the coulter counter analysis of crab meal; Dr. Joe Loesch for help with statistical analysis; Janet Walker, Maxine Butler, and Ruth Hershner of Word Processing for production of the final manuscript. I especially want to thank my parents for their considerable moral support and understanding throughout the course of my graduate studies. This study was funded in part by the Virginia Sea Grant College Program.

LIST OF TABLES

LIST OF TABLES (continued)

LIST OF FIGURES

ABSTRACT

Nursery culture of the juvenile hard clam, Mercenaria mercenaria (Linnaeus), a necessary step in commercial clam culture, is not considered economically feasible by many workers. This is largely due to the costs involved in supplying algae as the primary food source. An inexpensive, artificial food which could partially replace cultured algae would greatly reduce food costs.

Commercially available crab meal, a by-product of crab picking houses, was tested as a supplemental food with various sizes of juvenile hard clams. Feeding experiments were conducted in the summer of 1982 and from July to December in 1983 to determine optimum feeding methods and rates. In each experiment, both control and crab meal fed groups received filtered seawater at flow rates which contained enough natural food to support clam maintenance activities. Supplemented groups received crab meal at different rations proportional to total clam live weight. Various experiments tested methods of crab meal preparation. These included different sieve sizes of crab meal and autoclaving. Also different solutions were mixed with crab meal before delivery to trays. Growth was evaluated as the increase in shell height, and live, dry, and ash weights.

In all experiments, significantly greater increases in clam shell height and weight were observed in supplemented clams compared to control clams when crab meal was fed in proper amounts. Increases in clam shell height in control and crab meal fed clams were not directly correlated with seawater flow rates or chlorophyll *a* levels. There appeared to be a direct relationship between the percent increase in shell height and crab meal ration at optimum feeding rates. Optimum feeding rates for smaller clams (4-6 mm) were crab meal rations 20-24% of total clam live weight per day, and for larger clams (7-9 mm) rations were 11-15% of clam live weight per day. Crab meal which was sieved through 100 or 134 micron mesh, autoclaved, and mixed with 25 micron filtered seawater produced the greatest increases in clam weight and shell height. This study represents the first successful feeding of an artificial food to juvenile hard clams in a flow-through seawater system. These experiments, conducted under conditions similiar to those in commercial nurseries, indicate the potential for the use of crab meal in commercial nurseries as a partial replacement for cultured algae.

THE USE OF CRAB MEAL AS A SUPPLEMENTAL FOOD FOR JUVENILE HARD CLAMS (*MERCENARIA MERCENARIA*)

INTRODUCTION

Techniques necessary for the successful culture of the hard clam, Mercenaria mercenaria (Linnaeus), have existed for some time, yet commercial clam culture makes only a small contribution to the total annual clam landings (NMFS 1981). An overall decline in landings accompanied by an increased demand, however, creates an impetus for greater production of cultured clams. A major problem preventing commercial growers from greatly increasing their production involves the cost of operating clam nurseries. Shellfish hatcheries have not proven to be economically viable (Ryther 1981). The main reason for the economic failure of nurseries is the cost of feeding juvenile clams cultured algae as the main source of food (Epifanio 1976; McHugh 1981). Algal production is expensive due to the high costs involved in the filtration and sterilization of large volumes of seawater, the price of complex media, the energy for heating, cooling, and illuminating indoor cultures, and the necessary hardware and manpower (Persoone and Claus 1980). The cost of growing unicellular algal cultures as a food source for larvae is not prohibitive because of the relatively small amounts of algae required. However, much larger quantities are required to grow clam seed to juveniles with a shell height of 8-10 mm, a size suitable for planting in field plots. Juveniles of this size are less vulnerable to predators and thus have an increased chance of survival when planted in the field (Wallace

planted in the field (Wallace 1955; Mackenzie 1977; Castagna and Kraeuter 1981). Most hatchery operators pump large quantities of ambient seawater to post-set clams allowing the clams to utilize natural phytoplankton, bacteria, dissolved organics etc. for food. The electricity required to pump large volumes of water containing enough food for clam survival and growth can be costly.

Ultimately the best alternative to cultured algae as a primary food source would be the use of a nutritionally complete, formulated diet. This would enable a clam nursery system to become a profitable intensive culture operation. At present the complete nutritional requirements of filter-feeding bivalves is unknown (Langdon 1983a). Therefore a nutrionally complete artificial diet, a characteristic of high profit intensive culture systems, is not yet available.

A number of common foodstuffs and formulated diets have been tested on bivalves, mainly the oyster Crassostrea virginica, with varying success (Haven 1965; Gillepsie et al. 1966; Chanley and Normandin 1967; Dunathan et al. 1969; Harleston 1971; Castell and Trider 1974; Willis et al. 1976; Turgeon and Haven 1978; Epifanio 1979; Trider and Castell 1980; Ingle et al. 1981; Nell and Wisely 1983, 1984; Nell 1985). Carbohydrate rich sources such as cornstarch, cornmeal, dextrose, and wheat flour have been used to enhance growth. Haven (1965) found that cornstarch increased oyster meat weight but did not influence shell growth. Harleston (1971) fed cornstarch to adult mussels and clams and found that increases in the glycogen content of meats varied with season. The glycogen content of adult clams increased only with the feeding of cornstarch in the fall. Similarly, Turgeon and Haven (1978) showed that the effect of

cornstarch supplements on adult oysters varied with season. Oysters fed cornstarch during the fall and spring had higher glycogen levels than oysters normally found in natural surroundings. In their study cornstarch did not influence shell height and underwater shell weight during any season, which agreed with the earlier data of Haven (1965). Willis et al. (1976) fed cornmeal to adult oysters and observed an increase in glycogen content although an inverse relationship existed between oyster size and glycogen content after two and four weeks of feeding cornmeal. Cornmeal was also used successfully to fatten or increase the glycogen content of adult oysters in a commercial pilot plant operation (Ingle et al. 1981). During seasons when the condition of oysters is poor, the quality of meats was improved by the supplemental feeding of cornmeal in a few weeks.

Formulated artificial diets rather than individual food stuffs have also been fed to bivalves. Castell and Trider (1974) fed formulated diets with varying types and amounts of lipids to oysters. They observed differences in growth rate, meat weight, and glycogen content in response to diets containing different concentrations of cornstarch and different lipid types. Overall growth, however, did not approach that observed in oysters held in natural conditions. Nell and Wisely (1984) fed adult Sidney rock oysters, Saccostrea commercialis, artificial diets composed mainly of wheat starch and Pruteen, a bacterial protein. They found higher oyster meat glycogen contents in oysters fed this artificial diet compared to controls which received only flowing filtered seawater. Nell (1985) found increased glycogen content and condition index in adult S. commercialis fed artificial diets composed mainly of wheat starch with

Pruteen or the yeast Candida utilus. Most experiments using artificial foods as the main food source for adult bivalves have concentrated on fattening, i.e. increasing the glycogen content of adult bivalves. Other studies have been concerned with the use of artificial foods for bivalve larvae (Chanley and Normandin 1967; Lubet 1971; Masson 1977; Chu et al. 1982; Langdon 1983b).

Few studies have been performed on the use of non-algal foods or formulated artificial diets with juvenile bivalves (Lubet 1972; Gabbott et al. 1976; Murken 1976; Epifanio 1979; Langdon and Waldock 1981; Urban and Langdon 1984; Langdon and Bolton 1984; Langdon and Siegfried 1984). In most cases the artificial diet produced the best growth when it was fed with cultured algae. Murken (1976) fed fish processing wastes to juvenile mussels, Mytilus edulis alone and in combination with cultured algae and silt. When fish processing wastes served as the sole food source, a decrease in dry tissue occurred along with considerable mortality. The best growth occurred when mussels were fed combinations of fish processing wastes, cultured algae, and silt. Epifanio (1979) fed varying combinations of yeast and cultured algae to several species of juvenile bivalves. He found that it was possible to substitute up to 50% of the cultured algae in the diet with yeast and obtain growth comparable to a 100% cultured algal diet. Langdon and Bolton (1984) fed juvenile oysters encapsulated artificial diets. Oysters showed only minimal growth on the artificial diets, significantly less than the growth resulting from 100% cultured algae.

An alternative to cultured algae or a nutritionally complete formulated diet is the use of a supplemental food, which would enhance

growth and supply only part of the required nutrients. Supplemental feeding is often found in low intensity or extensive production systems, where natural foods are relied upon to meet all of the nutritional requirements of the cultured organism. More often a supplemental food is based on an inexpensive agricultural or processing by-product that is available in large quantities and inexpensive for cost effectiveness.

A potential supplemental food for juvenile clams is crab meal, a by-product of blue crab (Callinectes sapidus) picking operations. It is defined as the undecomposed ground, dried waste of the crab which contains the shell, viscera, and part or all of the flesh. Crab meal is inexpensive and readily available in quantities sufficient to be used in a clam hatchery operation (Murray and DuPaul 1981). Dunathan et al. (1969) fed crab meal to adult oysters and found that crab meal alone did not increase glycogen content or shell growth. When crab meal was fed with cornmeal in a 1:1 mixture, increases in glycogen, underwater total weight, and razor edge shell growth were observed when compared to unfed controls. These results, however, were not statistically analysed.

The following experiments were conducted to determine whether crab meal alone could serve as a supplemental food for juvenile hard clams. The previous experiments cited above have been largely concerned with increasing meat weights and/or glycogen content in adult bivalves. Increased tissue and shell size are important in juvenile clams for subsequent planting in field plots at a size less vulnerable to predators. The use of a supplemental food which would

promote overall growth of juvenile clams at a lower cost than cultured algae would reduce the costs of rearing larger size seed.

MATERIALS AND METHODS

Experiments were conducted at the Virginia Institute of Marine Science Eastern Shore laboratory during the spring and summer of 1982 and from July to December 1983. Hatchery-reared juveniles of Mercenaria mercenaria were obtained from selected broodstock clams which were spawned and reared in the same year for all experiments except in Experiment 2 (see page 16) which utilized clams from the previous year. Clams were held in raceways with flowing seawater filtered through a 50 micron GAF dacron bag prior to each experiment. Uniformly sized individuals with mean differences in shell height no greater than 0.5 mm were used in each experiment. Clams were randomly allocated to control and crab meal fed groups plus an additional group for determination of initial weights and shell height. Six experiments were designed to test the effect of the supplemental feeding of crab meal on the growth of different sizes of juvenile clams.

Crab meal was obtained from RCV Seafood, Moratico, Virginia. Table 1 shows the proximate analysis of crab meal. The amino acid content of crab meal before and after sieving through a 1mm screen is shown in Table 2. Crab meal sieved through a 1mm screen was crushed in a lapidary tumbler with carborundrum stones from a ball mill for five days. This ground crab meal was sieved through a 100 micron Nitex^{R} screen, autoclaved at 115.5^{o}C and 10 psi for 1 hour, dried in a mechanical convection oven at 60^{o}C for one hour, and later stored in a

TABLE 1. Proximate analysis of crab meal.[+]

Substance	Unsieved Crab Meal	Sieved Crab Meal
Protein %	38.0	42.3
Acid Detergent Fiber %	17.5	12.4
Ash %	3.8	3.5
Ether Extractable Fats %	4.2	6.6

TABLE 2. Amino Acid Profiles of Crab Meal (dry wt. basis).[+]

	Content (mg/g)	
Amino Acid*	Unsieved Crab Meal	Sieved Crab Meal
Lysine	11.226	14.458
Histidine	4.829	6.117
Ammonia	3.245	4.004
Arginine	13.135	16.224
Aspartic Acid	19.251	22.448
Threonine	8.747	9.792
Serine	9.083	9.740
Glutamic Acid	28.857	33.101
Proline	14.206	17.412
Glycine	10.593	12.757
Alanine	9.809	11.903
Half Cystine	-	0.742
Valine	10.098	11.291
Methionine	9.959	11.171
Isoleucine	9.859	10.260
Leucine	16.577	17.843
Tyrosine	8.924	9.756
Phenylalanine	9.121	10.474

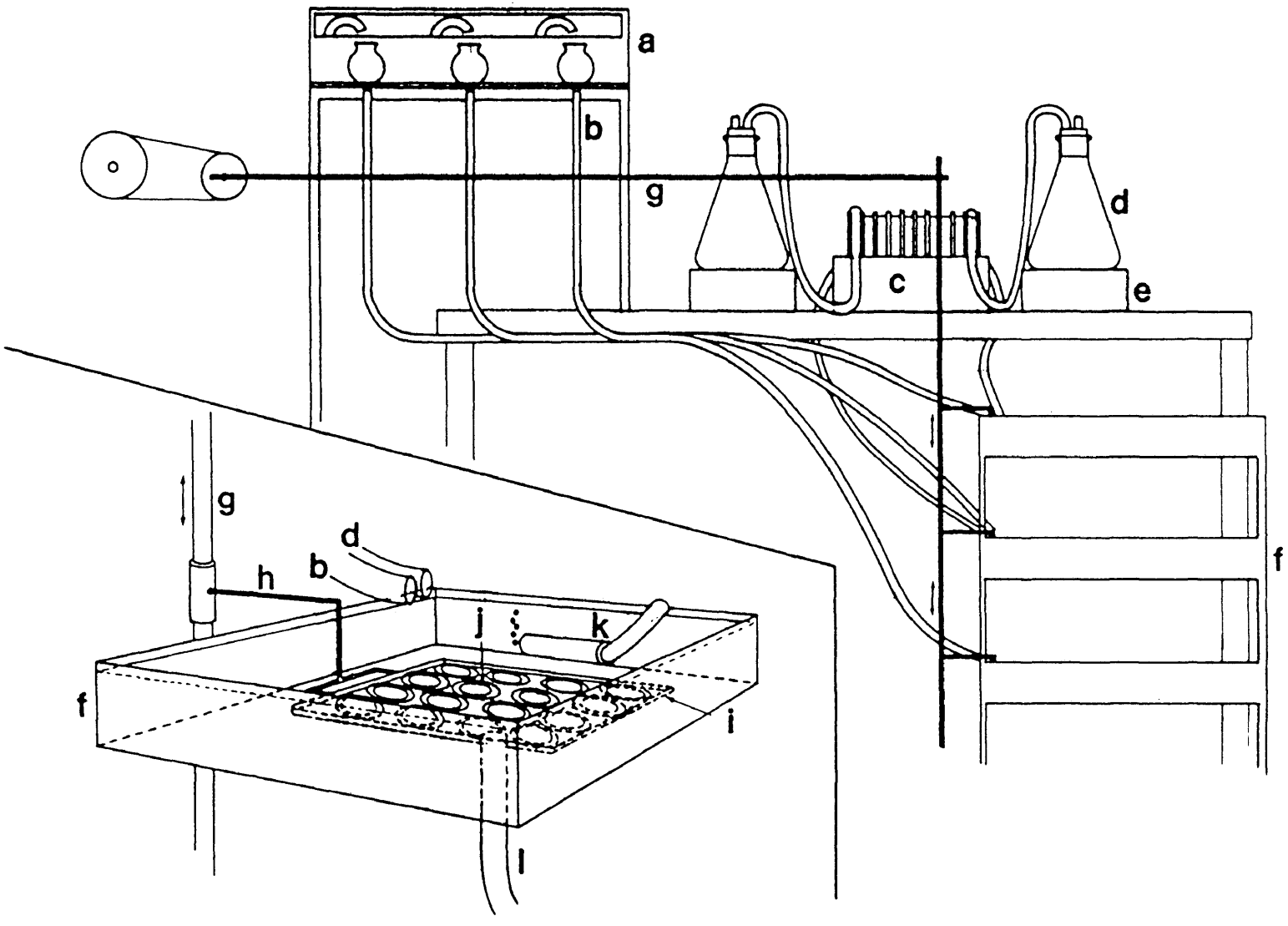
*The samples were hydrolyzed for 24 hours with hydrochloric acid.

[+]The proximate analysis and amino acid profiles of crab meal were determined by Dr. George J. Flick and Deborah Holloway of the Department of Food Science and Technology, Virginia Polytechnic Institute and State University.

dessicator at room temperature 22-26^{o}C. Rations of crab meal sufficient for two and three day periods were weighed on a H34 Mettler balance. Coulter counter analysis showed that 80% of the particles were less than 40 microns by volume. The mean particle size by population count was 20 microns. Rations were added to 4 l of 25 micron filtered seawater and mixed in a blender. Each crab meal suspension was poured into a 5 l Erlenmeyer flask which had a magnetic stirrer to keep the crab meal particles in suspension. Flasks were fitted with no. 10 rubber stoppers with two holes--one hole fitted with a short piece of glass tube to vent the solution and the other fitted with a glass tube connected to surgical then tygon tubing leading to a peristaltic pump. A variable speed Manostat R cassette peristaltic pump (Model no. 72-500) continuously delivered the food solution to the trays at 70 ml hr^{-1}.

Figure 1 shows the various components of the experimental apparatus. A single tray (61 X 50 X 7 cm) held each group of clams. Each tray had a metal rack which supported a screen holding sixteen containers. These containers were made from sections of PVC pipe (5 cm diameter, 1.5 cm height) with 1.5 mm mesh glued on to form a bottom. The metal rack support was joined at the side to a vertical bar which was connected to a motordriven chain and sprocket device to move the rack vertically in the tray at a rate of once every 8 minutes. This movement in the tray helped provide water circulation to the clams. Seawater filtered through a 50 micron core filter flowed into a headtank (with an overflow) which contained outlets to tubing leading to each tray. Seawater flow rates were adjusted by means of a screw clamp which varied the flow rate of water to thistle

Figure 1. Schematic design of experimental set-up for crab meal supplemental feeding experiments. a. headtank, b. seawater lines, c. peristaltic pump, d. crab meal solution, e. stirrer, f. tray, g. vertically moving metal rod, h. support rack, i. screen, j. clam container, k. air stone.


a
b
g
d
c
e
f
g
d
h
b
j
k
f
i
l

tubes connected to the surgical tubing leading to each tray. A water depth of 6 cm was always maintained in the trays. Airstones placed in the trays near seawater and food flow inlets helped keep food particles in suspension. Trays were drained by an outlet placed diagonally across the tray opposite the seawater inlet. The support structure holding the trays plus the food flasks were held in a controlled temperature room at 18^{o}C. A 12 hour photoperiod was maintained.

Before starting each experiment clams were sieved through appropriate size screens into one group of similiar size clams. Uniformly sized individuals with mean differences no greater than 0.5 mm were used in each experiment. Experimental groups plus an additional group for initial sacrifice were randomly allocated from this group. Live, dry, and ash weights were determined on the sacrificed group plus the shell heights (hinge to lip measurement) of 100 randomly chosen clams were measured. Heights were measured using Vernier calipers to the nearest 0.1 mm in Experiments 1 and 2 (p. 15 - 16). In the remaining experiments 3-6, a dissecting microscope fitted with an ocular micrometer was used to measure the shell height of photocopies of the clams (Haines 1973). Live weights were determined after clams had been air dried for one hour. Dry weights were taken after clams were dried in a mechanical convection oven at 60^{o}C for one week and placed in a desiccator for two hours. Ash weights were determined after clams were incinerated for four hours at 500^{o}C in a muffle furnace. All weights were measured to the nearest 0.1 mg on a H34 Mettler balance.

The remaining groups of clams were randomly assigned to one control and one or more treatment groups. Treatments involved specific amounts and preparation techniques of crab meal as a supplemental food. All groups received 50 micron filtered seawater at the same flow rate. Preliminary experiments determined that this flow rate contained at least enough natural food to serve as a maintenance ration or the amount of food necessary to maintain a constant weight (Winter and Langton 1976). Crab meal was fed at a predetermined percentage of total clam live weight. Throughout the experiment as clam weight increased, rations were readjusted to maintain the initial feeding rate. Table 3 gives the various seawater flow rates, crab meal rations, and clam sizes involved in each experiment.

Seawater flow rates were measured daily and adjusted if necessary after the core filter had been changed. Water temperatures in the trays were recorded and adjusted where required with electric immersion aquarium heaters. Twice a week containers were rinsed to remove debris from the screened bottoms. Once a week the trays were drained, rinsed with fresh water, and refilled with 50 micron filtered seawater at the same temperature of the previous water. Chlorophyll *a* samples were taken daily from the head tank serving the experimental trays for later analysis. Salinity values were obtained from ongoing daily measurements taken at Wachapreague.

Final clam live, dry, and ash weights plus shell height were used to evaluate the effect of crab meal on clam growth. One-way analysis of variance (ANOVA) was performed to discriminate differences between the final weights or shell heights of the different treatments at a level of inference of $\alpha = 0.01$ (Sokal and Rohlf 1981). Homogeneity of

TABLE 3. Protocol of crab meal supplemental feeding experiments 1-6 including temperature, chlorophyll *a*, flow rates, clam sizes, crab meal rations, and various treatments.

Exp. No.	Date	Mean temp. range (°C)	Mean chlorophyll + range ($\mu g\ l^{-1}$) ($\pm$ 1 SD)	Seawater flow rates ($ml\ min^{-1}$)	Initial Clam height (mm) ($\pm$ 1 SD)	Crab meal % clam live wt.	Treatments
	1982						
1	8/18-9/17	20.0 19.5-25.0	not taken	106	5.5	18	1. ration mixed with 25 µm filtered seawater
						0	2. control
	1983						
2	7/12-8/11	23.0 19.0-24.0	not taken	180	9.4 (0.6)	16	1. ration mixed with 25 µm filtered seawater
						11	2. same as above
						11	3. same as above, but 53 µm crab meal
						11	4. same as above, crab meal not autoclaved
						0	5. control
3	8/11-9/11	19.3 17.5-20.7	4.34 (5.51) 0.32-14.66	35	4.9 (0.4)	24	1. ration mixed with 25 µm filtered seawater
						0	2. control
4	8/1-8/22	19.0 17.5-20.7	5.22 (4.08) 0.56-13.87	30	4.0 (0.3)	30	1. ration mixed with 25 µm filtered seawater
						30	2. ration mixed with brine solution
						0	3. control
5	9/30-10/29	18.3 15.5-18.5	2.39 (1.88) 0.42-7.33	156	6.9 (0.4)	20	1. ration mixed with 10 µm filtered seawater
						20	2. ration mixed with brine solution
						0	3. control
6	11/10-12/10	19.0 14.5-24.0	2.52 (1.52) 0.40-7.23	96	7.1 (0.3)	15	1. ration mixed with 25 µm filtered seawater
						0	2. control

variance was determined using Cochran's and Bartlett's tests. Because of the robustness of the F test and the very large and nearly equal number of individuals in each treatment, one-way ANOVA was performed on untransformed data even when there was heterogeneity of variance among the groups according to Bartlett's and Cochran's C tests (Sokal and Rohlf 1981). The Student-Newman-Keuls multiple range test was used to determine differences between means at a level of inference of $\alpha = 0.05$, where a significant value for the F test was found.

Experiment 1

Hatchery reared juvenile clams were sieved through a 5 mm screen and caught on a 4 mm screen. One hundred clams were chosen for initial sacrifice and two groups of 400 clams each were chosen for the experimental groups. Clams ranged in shell height from 5-6 mm and had a mean wet weight of 67.5 $\pm$ 5.0 mg ($\pm$ 1 SD).

Crab meal was prepared as described above except that it was sieved through a 35 micron screen instead of a 100 micron screen after the crab meal was ground in the lapidary tumbler for one week. Crab meal rations were 18% of clam live weight which equalled 5 g crab meal per tray per day. Crab meal was mixed with 5 micron GAF bag filtered seawater for the first week and with 25 micron filtered seawater for the remainder of the experiment.

Initial sacrifice of 100 clams was performed and wet, dry, and ash weights were taken. The remaining two groups of 400 clams were placed in trays one and two. Sixteen containers each holding 25 clams were put in both trays. Each tray received 50 micron filtered seawater at a flow rate of 106 ml min^{-1}. The control group in tray 1 received only the natural food contained in the filtered seawater

while group 2 received crab meal in addition to the naturally occurring food.

Twenty-six randomly selected clams were removed from each group, measured from hinge to lip (shell height) with vernier calipers, weighed, marked, and returned to their original positions in each tray. Live weight and shell height were measured on these individual clams again on days 5, 10, 15, and 30 of the experiment. At the completion of thirty days, all clams were sacrificed and live, dry, and ash weights were obtained as stated earlier.

Salinity ranged from 29.0-32.0 ppt with a mean of 30.7 ppt. No chlorophyll *a* measurements were made. Table 3 contains water temperatures.

Experiment 2

Hatchery-reared juvenile clams used in this experiment had been spawned in the previous summer and were considered "slow growers". Approximately 1,300 clams were randomly selected from this group. The mean shell height was 9.4 mm $\pm$ 0.6 mm and the mean live weight 313.3 mg $\pm$ 21.0 mg. Clams were divided into five groups containing 240 clams each plus an additional group of 125 clams for sacrifice to determine initial weights and heights. The five groups of clams were randomly allocated to one control and four treatment groups.

Each group of clams was placed in a tray with a flow rate of 180 ml min^{-1} of 50 micron filtered seawater. Treatments involved comparisons on crab meal preparation--whether it was autoclaved or not, particle size (53 or 100 micron), and concentration (0, 11, and 16%) (Table 4). Crab meal was fed at amounts 11 or 16% of the total clam live weight per tray which equalled 8 and 12 g per day

TABLE 4. Explanation of the various treatments for experiment 2 concerning crab meal concentrations, particle size (sieve size used with crab meal), and whether autoclaved or not.

Tray No.	Crab Meal Concentration (grams/day)	Sieve Size (microns)	Autoclaved (1 hr. at 115°C)
1	12	100	yes
2	8	100	yes
3	8	53	yes
4	8	100	no

respectively. Throughout the experiment as clam weight increased, rations were readjusted to maintain the proper amount based on live weight. Random samples of 75 clams from each group were collected and measured every six days.

Salinity ranged from 32-34 ppt with a mean of 32.9 ± 0.6 ppt. No chlorophyll *a* measurements were made. Water temperature is given in Table 3.

Experiment 3

Hatchery-reared juvenile clams, produced in the spring 1983 spawning of selected broodstock, were sieved through a 4 mm screen and caught on a 3 mm screen. Approximately 1,300 clams were randomly selected for the experiment-- two groups (400 each) for the treatment and control, and one group (400) for initial sacrifice. Clams had a mean shell height of 4.9 mm ± 0.4 mm and a mean live weight of 45.2 mg ± 4.3 mg.

The experimental design described for experiment 2 was used in this study. Each tray held 400 clams, with 25 clams placed in each container. Seawater, filtered through a 50 micron core filter, flowed through each tray at a rate of 35 ml min^{-1}. The supplemented tray received crab meal at 70 ml hr^{-1}. Partial water changes were carried out weekly. The clams were rinsed off with fresh water twice a week to remove debris from the containers.

Crab meal was prepared as stated earlier-- being sieved through a 134 micron Nitex^{R} screen and autoclaved at 115.5^{o}C and 10 psi for one hour. Crab meal was fed at a concentration 24% of the total clam live weight which equalled 4.3 g of crab meal per day initially. Two and three day rations were blended with 4 l of 25 micron bag filtered

seawater and added to 5 l Erlenmeyer flasks. Four randomly selected containers of clams were removed from each tray every six days and live weights were measured. Crab meal rations were then readjusted according to any change in clam weight.

Four hundred clams were sacrificed for initial weight determinations. Photocopies of these clams were made for later measurement of shell height (Haines 1973). All weights were determined as described in experiment 1. At the completion of this experiment all clams were sacrificed, and live, dry, and ash weights were measured. All clams were photocopied and shell heights were measured on 100 randomly selected clams from each group.

Chlorophyll *a* samples were taken daily during high water in the afternoon or evening from the head tank containing the filtered sea water which supplied the trays. Samples were filtered through a Whatman GF/F filter and placed in blackened test tubes containing 8 ml of acetone-DMSO-water (9:9:2) extraction solutions (Haywood and Webb 1981). Samples were stored in a refrigerator at 8^{o}C until measured for fluorescence in a G.K. Turner Associates model 111 fluorometer fitted with a standard door.

Salinity ranged from 32.0-32.5 ppt with a mean of 32.4 ppt. Ph of the trays varied from 7.4-7.8 with a mean of 7.6 during the experiment. Water temperature and chlorophyll *a* are given in Table 3.

Experiment 4

Randomly selected juvenile clams sieved through a 3 mm screen and caught on a 2 mm screen were split into four groups of 480 clams each. One group was sacrificed and initial weights were obtained. Photocopies of these clams were made for initial shell height

measurements. The remaining groups were randomly allocated to one control and two groups to receive crab meal. Clams had a mean live weight of 26.1 mg ± 0.95 mg and a mean shell height of 4.0 mm ± 0.29 mm.

The same experimental design was used as in experiments 2 and 3. Each container held 30 clams making a total of 480 clams per tray. All three trays received seawater filtered through a 50 micron core filter at a rate of 30 ml min^{-1}. Crab meal was prepared as stated in experiment 3. Crab meal was fed to trays one and two at 30% of the total clam live weight which equalled 3.8 g per day initially. Tray 1 held clams which were given crab meal mixed with 25 micron bag filtered seawater. Tray 2 received crab meal mixed with a brine solution of fresh water and 280 g of rock salt. In tray 3 control clams received only the natural food contained in 50 micron filtered seawater. Each week trays were rinsed with fresh water and refilled with filtered seawater at the same temperature as the previous tray water. Individual clam containers were rinsed with fresh water twice a week to remove debris clogging the screens. Any dead clams were replaced with clams of the same size. Every five or six days four containers were randomly selected from each tray and live weights were obtained. Chlorophyll *a* samples were taken daily near high water in the afternoon or evening. Temperatures were recorded daily from each tray.

Plankton counts were made periodically for each tray and the head tank supplying the trays using epifluorescent microscopy. These slides were made following the procedure of Haas (1982). Samples were taken from each tray in a similiar location. Samples of 2 ml were

taken from the control tray or head tank and 1 ml from the trays which received crab meal. Water samples were added to a Millipore filter apparatus containing a 0.2 micron Nuclepore filter which had been prestained in irgalen black placed on top of a Millipore HA filter. Proflavin (0.33% in distilled water) was then added to the sample (20 µl per ml) and gently agitated. After 2 minutes gluteraldehyde (6.0% W/V in 0.22 micron prefiltered seawater) was added (50 µl/ml) to the filter apparatus and the sample was again agitated. After two minutes, a vacuum (less than 5 cm Hg) was applied and the top filter was removed from the apparatus when the meniscus had disappeared, while the vacuum was still being applied. The filter was placed, top side up, on a slide containing one drop of immersion oil. Another drop of immersion oil was added plus a cover slip. Slides were stored in a freezer at -15°C.

Salinity ranged from 31.0 to 32.5 with a mean of 32.1 ± 0.4 during the experiment. Water temperature and chlorophyll *a* are given in Table 3.

Experiment 5

Randomly selected hatchery-reared juvenile clams sieved through 6 mm screen and caught on a 5 mm screen were split into four equal groups of 400 clams each--one group for initial sacrifice and four groups for different treatments. Clams had a mean height of 6.9 mm ± 0.4 mm and a mean weight of 156.6 mg ± 6.9 mg.

The same experimental design was used as described in experiment 1-4. Each tray held 400 clams. Fifty micron filtered seawater flowed through the trays at a rate of 156 ml min^{-1}. Aquarium heaters were placed in each tray to regulate water temperature. Trays were rinsed

out twice a week with fresh water and refilled with 50 micron filtered seawater at the previous tray temperature.

Crab meal was prepared as stated in experiments 3 and 4. Supplemental feeding of crab meal was at 20% of total clam live weight in trays 1 and 2. Tray one received crab meal mixed with 4 l of 10 micron filtered seawater. Tray 2 received crab meal rations mixed with fresh water and 280 g of salt to make a brine solution. Occasionally, if crab meal solutions had drained too quickly into the trays, approximately 500 ml of 10 micron filtered seawater or fresh water (depending on the treatment) would be added to the remaining food in the flask and delivered to the trays as usual.

Chlorophyll *a* samples were taken daily near high water in the afternoon or evening as described in experiments 3-5. Epifluorescent slides were made periodically for plankton counts.

The mean salinity was 29.6 $\pm$ 1.3 ppt and ranged from 28-31.5 ppt. Ph varied from 7.8-8.0 with a mean of 7.9. Table 3 gives chlorophyll *a* and water temperatures.

Experiment 6

Hatchery-reared juvenile clams which fell through a 6 mm sieve and were caught on a 5 mm sieve were randomly split into three groups of 400 clams each. One group was sacrificed for initial weight and height determinations while the other groups were split into one control and one crab meal fed group. Clams had a mean shell height of 7.1 mm $\pm$ 0.3 mm and a mean weight of 154.8 mg $\pm$ 8.1 mg.

Clams were placed in the same experimental design as described above. All trays received 50 micron core filtered seawater at 96 ml min^{-1}. Each tray contained 400 clams, 25 clams per container. Trays

were rinsed completely once a week with fresh water and refilled with 50 micron filtered seawater at the same temperature as the previous water. Trays received partial seawater changes every three days. An aquarium heater was placed in each tray to keep the seawater temperature within an acceptable range.

Crab meal was prepared as stated in experiments 3-5. Two and three day rations were mixed with 4 l of 25 micron filtered seawater. Occasionally when crab meal solutions had drained too quickly into the trays, approximately 500 ml of 25 micron filtered seawater would be added to the remaining food in the food flasks and fed as usual. Clams in tray 1 were fed crab meal at 15% of their live weight which initially was 9.3 g per day per tray. Clams in tray 2 received only the natural food contained in the 50 micron filtered seawater which all the trays received.

Chlorophyll *a* samples were taken as stated earlier. Random samples of four containers of clams were taken weekly from each tray and live weights were obtained. Epifluorescent slides for plankton counts were made periodically.

Mean salinity was 30.0 $\pm$ 1.0 ppt and ranged from 26.5-32.0 ppt. Tray water temperatures were maintained at 19.0^{o}C throughout the experiment with the use of heaters whereas the mean ambient water temperature was 10.0 $\pm$ 2.2^{o}C. Table 3 gives water temperature and chlorophyll *a*.

RESULTS

Experiment 1

The increase in live weight of randomly selected clams during the course of the experiment is shown in Figure 2. Clams fed crab meal at 18% cent of clam live weight per day showed greater increases in live, dry, and ash weights than the unsupplemented control clams which received filtered seawater only, at the conclusion of 30 days of feeding (Table 5). Increases in live and dry weights of crab meal supplemented clams were 2.5 times greater than those of control clams. One-way analysis of variance (ANOVA) showed that live, dry, and ash weights of crab meal fed clams were significantly greater than those of control clams ($P<0.01$) (Table 6).

Experiment 2

The final shell heights were greatest in treatment 2 (TRT 2) followed by TRT 3 both of which received crab meal at 11% of live weight (Table 7). Treatments 2, 3, and 4 (crab meal at 11% live weight) all resulted in significantly greater shell heights than treatment 1 (crab meal at 16% live weight) or the control (TRT 5) (Table 8). Little difference was found between the final shell height of the control (TRT 5) and the initial shell height (Figure 3). No significant difference was found between the final shell height of clams fed crab meal at 16% live weight and of control clams (SNK, $P<0.01$) (Table 8). Clams in TRT 2 showed the greatest increase in shell height (0.4 mm).

Figure 2. Change in live weight (grams) of crab meal supplemented and control clams during Experiment 1 (duration of experiment = 30 days). Each data point represents the mean of 26 randomly selected clams.

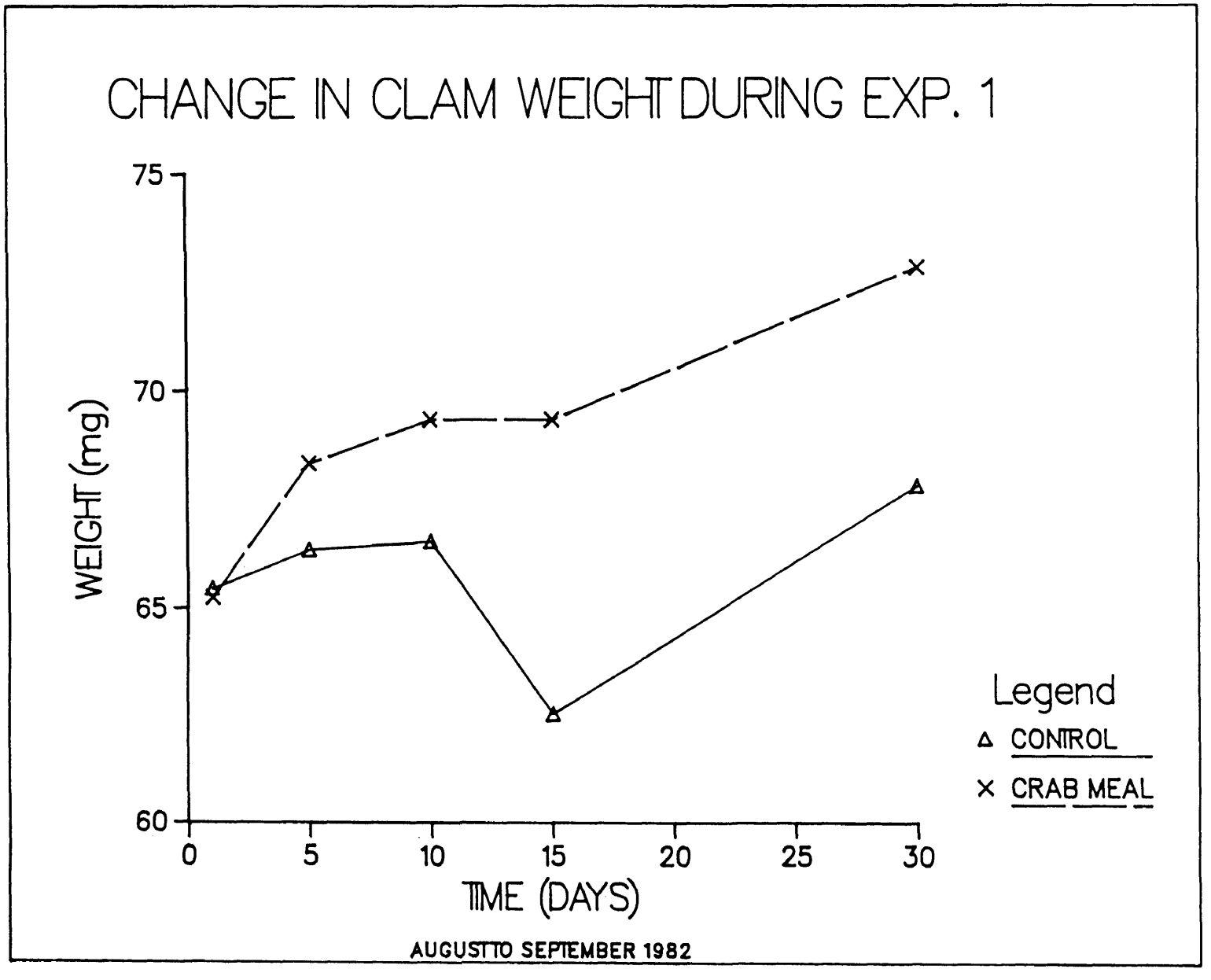

CHANGE IN CLAM WEIGHT DURING EXP. 1
75
70
65
60
WEIGHT (mg)
0
5
10
15
20
25
30
TIME (DAYS)
Legend
CONTROL
CRAB MEAL
AUGUSTTO SEPTEMBER 1982

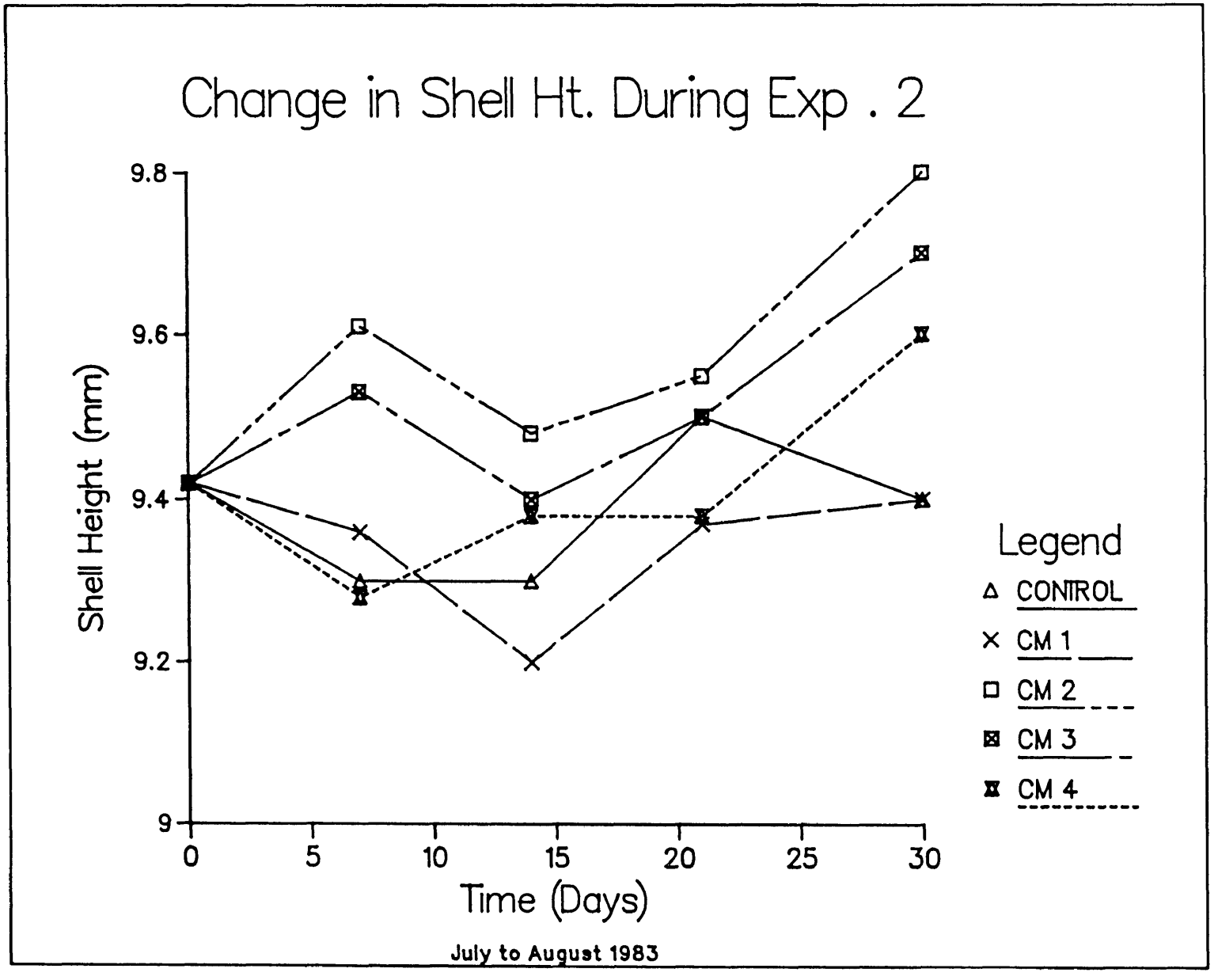
TABLE 5. Experiment 1. Initial and final mean live, dry, and ash weights (mg) of clams held in 106 ml min^{-1} flowing sea water with and without crab meal supplementation after 30 days (initial n=125, final n=400).

	Initial	Final	
		Crab Meal	Control
live weight (mg)			
$\bar{X}$	67.5	71.4	68.2
SD	3.8	5.5	5.8
dry weight (mg)			
$\bar{X}$	41.0	45.1	42.4
SD	4.3	2.7	3.5
ash weight (mg)			
$\bar{X}$	38.3	41.3	39.8
SD	3.4	3.0	3.4

TABLE 6. Experiment I. Comparison of mean final weights and shell heights of juvenile clams (mean ± 1 SD) held in trays of 106 ml min^{-1} flowing filtered seawater with and without crab meal supplementation (n=400 per tray, duration of experiment = 30 days). F ratios and probabilities are from one-way analysis of variance of final weights and heights.

	Treatments			
Final	crab meal (18% live wt.)	control	F Ratio	Probability
Live weight (mg)	71.4 (5.5)	68.2 (5.8)	61.208	0.000
Dry weight (mg)	45.1 (2.7)	42.4 (3.5)	134.099	0.000
Ash weight (mg)	41.3 (3.0)	39.8 (3.4)	44.279	0.000

Figure 3. Changes in shell height (mm) of crab meal supplemented (TRT's 1-4) and control (TRT 5) clams during Experiment 2 (duration of experiment = 30 days). Each data point represents the mean of 75 randomly selected clams.


Change in Shell Ht. During Exp . 2
Shell Height (mm)
9.8
9.6
9.4
9.2
9
0
5
10
15
20
25
30
Time (Days)
Legend
CONTROL
CM 1
CM 2
CM 3
CM 4
July to August 1983

One-way ANOVA revealed significant differences in clam growth depending on the ration size and methods of preparation employed when using crab meal as a supplemental food ($P<0.01$). Crab meal rations of 11% clam live weight resulted in significantly greater growth in clams than rations of 16% live weight or than in unsupplemented control clams (Table 8). Final live, dry, and ash weights of clams fed crab meal of the highest ration, treatment (TRT) 1, were significantly lower than the final weights of unsupplemented controls (SNK, $P<.01$) and than the other groups fed crab meal (Table 8). Clams fed this higher ration of crab meal showed little weight gain.

Smaller increments in growth parameters were seen when crab meal was fed without being autoclaved in TRT 4 compared to autoclaved crab meal (TRT 2 & 3) at the same ration (Table 7). Clams fed unautoclaved diets showed lower final live, dry and ash weights than unsupplemented control clams. In relation to crab meal particle size, clams fed 100 micron sieved crab meal showed little difference in weight compared to clams fed crab meal sieved through a 53 micron screen (TRT 3). No significant difference between the live weight of TRT 2 and 3 was found, although the dry and ash weights of TRT 2 clams were significantly greater than in TRT 3.

Overall, clams fed autoclaved crab meal at 11% of live weight, which was sieved through a 100 micron mesh, showed the greatest increase in growth parameters of all treatments. No significant difference was found concerning shell height between clams fed 53 or 100 micron sieved crab meal, but crab meal of the larger sieve size resulted in significantly greater ash and dry weights.

TABLE 7. Experiment 2. Initial and final live, dry, and ash weights (mg) and shell heights (mm) of clams held in 180 ml min^{-1} flowing seawater with and without crab meal supplementation after 30 days (initial n=125, others n=240).

	Initial	Final				
		1	2	3	4	5
		Crab Meal	Crab Meal	Crab Meal	Crab Meal	Control
live weight (mg)						
$\bar{X}$	313.3	337.7	353.3	353.0	336.7	353.9
SD	21.0	17.1	19.6	16.3	16.3	23.6
dry weight (mg)						
$\bar{X}$	202.3	214.6	238.3	233.1	224.2	234.8
SD	14.5	10.5	9.4	11.9	10.8	12.7
ash weight (mg)						
$\bar{X}$	191.0	196.4	219.1	216.9	203.9	214.3
SD	19.0	10.1	10.9	8.4	10.7	12.3
shell height (mm)						
$\bar{X}$	9.4	9.4	9.8	9.7	9.6	9.4
SD	0.6	0.6	0.6	0.7	0.7	0.8

TABLE 8. Experiment 2. Comparison of mean final weights and shell heights of juvenile clams (mean ± 1 SD) held in trays with 180 ml min^{-1} flowing filtered seawater with and without crab meal supplementation for 30 days (n=240 per tray). F ratios and probabilities are from one-way analysis of variance of final weights and heights. Student-Newman-Keuls (SNK) range tests show subsets of groups at the 0.05 level of significance.

Final	Treatments 1 Crab meal (16)%	2 Crab meal (11%)	3 Crab meal (11%)	4 Crab meal (11%)	5 Control	F ratio	Probability
Live weight (mg)	333.7 (17.1)	353.3 (19.6)	353.0 (16.3)	336.7 (16.3)	353.9 (23.6)	8.391	0.0000
Dry weight (mg)	214.6 (10.4)	238.3 (9.4)	233.1 (11.9)	224.2 (10.8)	234.8 (12.7)	137.029	0.0000
Ash weight (mg)	196.4 (10.1)	219.1 (10.9)	216.9 (8.4)	203.9 (10.7)	214.3 (12.3)	191.994	0.0000
Shell height (mm)	9.4 (0.6)	9.8 (0.6)	9.7 (0.7)	9.6 (0.7)	9.4 (0.8)	4.559	0.0013

Student-Newman-Keuls

Live weight	1	4	3	2	5
Dry weight	1	4	3	5	2
Ash weight	1	4	5	3	2
Shell height	5	1	4	3	2

Experiment 3

Crab meal supplemented clams showed increases in live, dry, and ash weight approximately 1.5 times greater than control clams fed solely on natural foods (Table 9). Clams fed crab meal at 24% live weight showed significantly greater final live, dry, and ash weights than control clams ($P<0.01$) (Table 10). Figure 4 shows the increase in weight of randomly selected clams during the course of the experiment. Shell heights were also significantly greater in crab meal supplemented than in control clams (ANOVA, $P<0.01$) (Table 10). Crab meal fed clams showed an increase in shell height five times greater than in control clams. An increase of 1.0 mm in 30 days is better than average for recorded growth rates in natural settings during August and September for this latitude (Ansell 1968). A neglible rate of mortality occured in this experiment (< 2%).

Experiment 4

Clams fed crab meal at 30% of live weight mixed with freshwater and rock salt solution (TRT 2) showed the greatest increase in shell height and weight (Table 11). Live, dry, and ash weights of TRT 2 were significantly greater than those of the control or TRT 1 (crab meal mixed with filtered seawater) (ANOVA, $P<0.01$) (Table 12). Figure 5 shows the increase in live weight of randomly sampled clams during the course of the experiment. Shell heights of both crab meal supplemented clams were significantly greater than control clam shell height (ANOVA, $P<0.01$).

Considerable mortality occurred in this experiment which tested the smallest sizes of clams (4.0 mm) with the largest crab meal ration. Clams of nearly the same size were always used to replace

Figure 4. Change in live weight (mg) of crab meal supplemented and control clams during Experiment 3 (duration of experiment = 30 days). Each data point represents the mean live weight of 100 randomly selected clams.

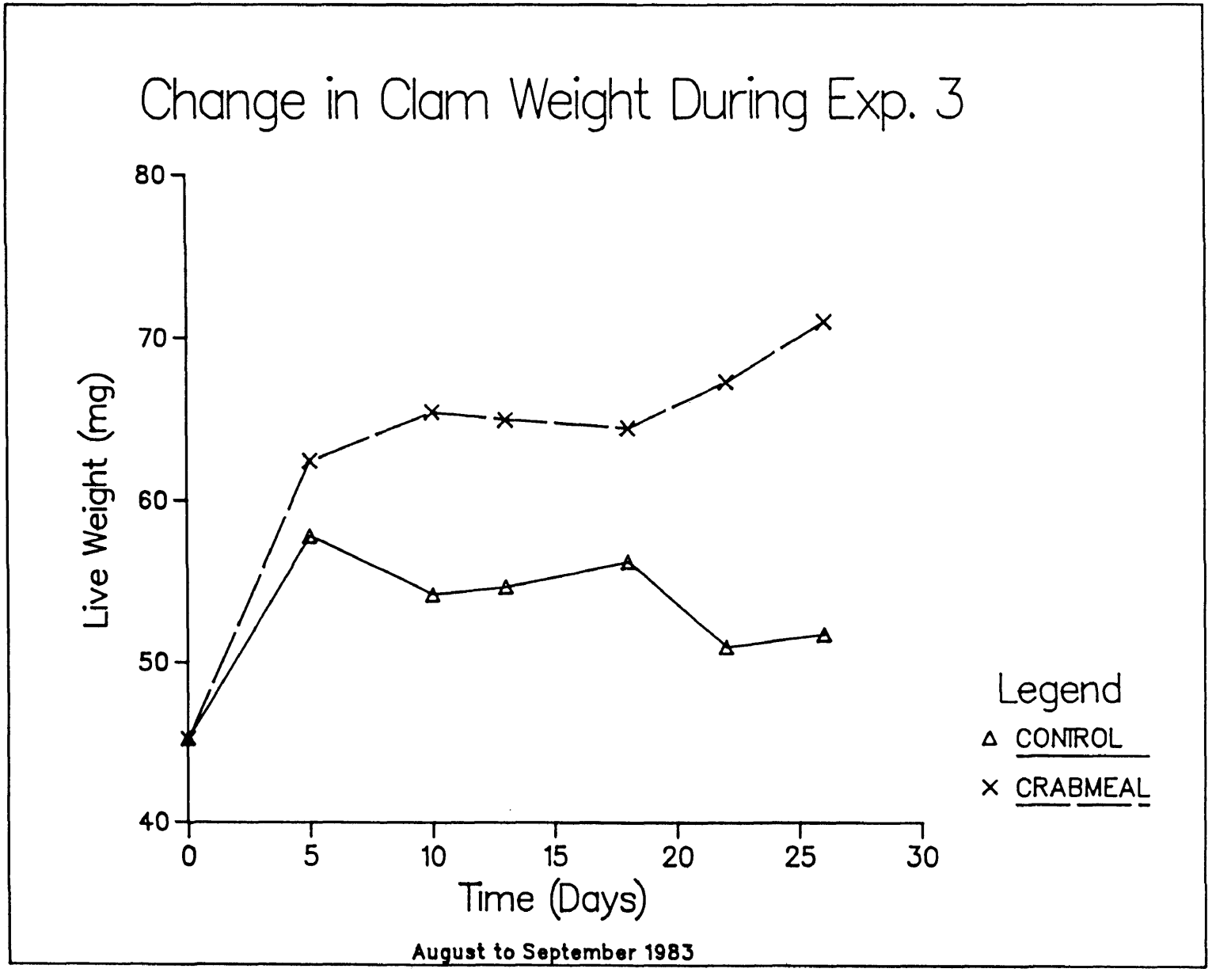

Change in Clam Weight During Exp. 3
Live Weight (mg)
80
70
60
50
40
0
5
10
15
20
25
30
Time (Days)
Legend
CONTROL
CRABMEAL
August to September 1983

TABLE 9. Experiment 3. Initial and final mean live, dry, and ash weights (mg) and shell heights (mm) of juvenile clams held in 35 ml min^{-1} filtered flowing seawater with and without crab meal supplementation after 30 days (n= 400 per group).

	Initial	Final	
		Crab Meal (24%)	Control
live weight (mg)			
$\bar{X}$	45.2	64.9	57.8
SD	4.3	6.8	9.3
dry weight (mg)			
$\bar{X}$	29.5	41.2	37.3
SD	3.3	4.2	6.3
ash weight (mg)			
$\bar{X}$	28.2	38.5	35.2
SD	1.7	3.7	5.9
shell height (mm)			
$\bar{X}$	4.9	5.9	5.1
SD	0.4	0.5	0.6

TABLE 10. Experiment 3. Comparison of mean final weights and shell heights of juvenile clams (mean ± 1 SD) held in 35 ml min^{-1} flowing filtered seawater with and without crab meal supplementation (n=400 per tray, duration of experiment = 30 days). F ratios and probabilities are from one-way analysis of variance of final weights and heights.

	Treatments			
	1	2		
Final	control	crab meal (24%)	F ratio	Probability
Live weight (mg)	57.8 (9.3)	64.9 (6.8)	143.278	0.000
Dry weight (mg)	37.3 (6.3)	41.2 (4.2)	101.936	0.000
Ash weight (mg)	35.2 (5.9)	38.5 (3.7)	85.309	0.000
Shell height (mm)	5.1 (0.6)	5.9 (0.5)	99.388	0.000

those which died. Besides being the smallest clams tested, crab meal was also fed at the highest concentration. Small clams are known to have higher mortality rates but increased bacterial levels due to the high concentration of crab meal was probably also a contributing factor. Higher mortality was seen in TRT 1 where crab meal was mixed with filtered seawater than in the control or in TRT 2.

Bacterial numbers in the trays receiving crab meal were greater in this experiment than in any others examined. Water samples from trays receiving crab meal had 2-3 times more bacteria and heterotrophic flagellates than the control tray. Samples from TRT 1 tray water, where crab meal was mixed with seawater, had at least two times the number of bacteria as TRT 2 tray water, where crab meal and brine solutions entered initially.

Experiment 5

Clams which received crab meal mixed with filtered seawater (TRT 1) showed greater increases in shell height than unsupplemented control clams (Table 13). TRT 1 clams fed crab meal at 20% live weight showed significantly greater dry and ash weights than TRT 2 clams which received the same ration of crab meal mixed with a brine solution (ANOVA, $P<0.01$) (Table 14). Clams receiving the crab meal and brine solution (TRT 2) showed significantly lower weights than control or TRT 1 clams. (SNK, $P<0.05$). Both crab meal fed clam groups showed significantly greater increases in shell height than unsupplemented clams (Table 13). There was no significant difference between the control and the TRT 1 group in terms of live weight.

During the course of the experiment large red blotches of bacteria appeared on tray 2. Clams were observed to feed less when

Figure 5. Change in live weight (mg) of crab meal supplemented and control clams during Experiment 4 (duration of experiment = 21 days). Each data point represents the mean of 120 randomly selected clams.

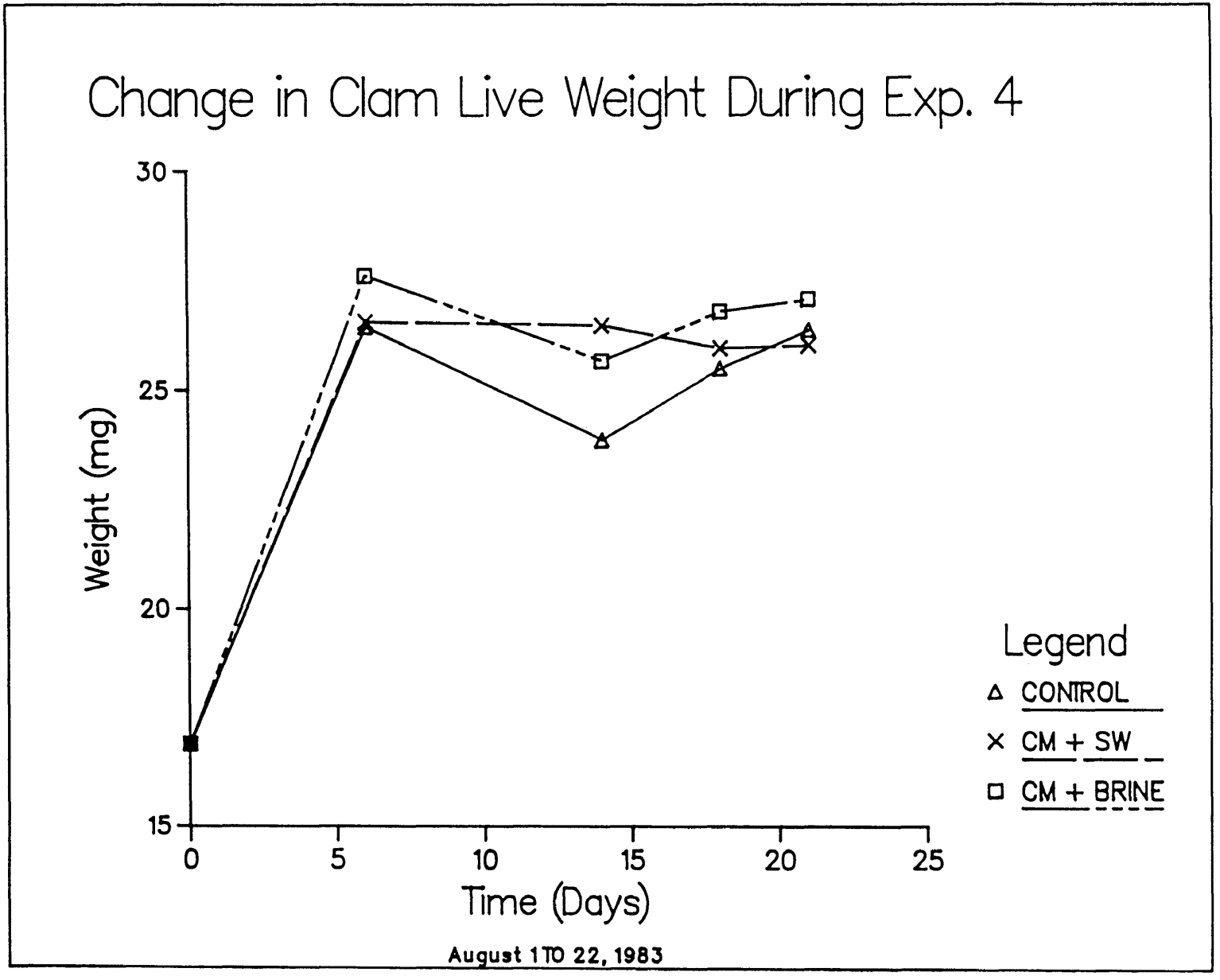

Change in Clam Live Weight During Exp. 4
30
25
20
15
Weight (mg)
0
5
10
15
20
25
Time (Days)
Legend
CONTROL
CM + SW
CM + BRINE
August 1 TO 22, 1983

TABLE 11. Experiment 4. Initial and final live, dry, and ash weights (mg) and shell heights (mm) of clams held in trays with 30 ml min^{-1} flowing seawater with and without crab meal supplementation (control) for 21 days (n=450 per tray).

	Initial	Final		
		1	2	3
		Crab Meal (30%)	Crab Meal (30%)	0 control
live weight (mg)				
$\bar{X}$	26.1	26.1	27.1	26.3
SD	1.3	2.0	1.4	2.1
dry weight (mg)				
$\bar{X}$	16.9	17.0	17.5	17.0
SD	1.0	1.2	0.9	1.0
ash weight (mg)				
$\bar{X}$	15.6	15.9	16.4	15.7
SD	0.8	1.1	0.6	1.0
shell height (mm)				
$\bar{X}$	4.0	4.2	4.1	4.0
SD	0.2	0.4	0.4	0.3

TABLE 12. Experiment 4. Comparison of mean final weights and shell heights of juvenile clams (mean ± 1 SD) held in trays with 30 ml min^{-1} flowing filtered seawater with crab meal supplementation for 21 days (n=450 per tray). F ratios and probabilities are from one way analysis of variance of final weights and heights. Student-Newman-Keuls (SNK) range tests show subsets of groups at a level of significance of α = 0.05.

	Treatments				
	1	2	3		
Final	Crab meal (30%)*	Crab meal (30%)*	Control	F ratio	Probability
Live weight (mg)	26.1 (2.0)	27.1 (1.4)	26.3 (2.1)	29.158	0.000
Dry weight (mg)	17.0 (1.2)	17.5 (0.9)	17.0 (1.0)	29.311	0.000
Ash weight (mg)	15.9 (1.1)	16.4 (0.6)	15.7 (1.0)	78.677	0.000
Shell height (mm)	4.2 (0.4)	4.1 (0.3)	4.0 (0.3)	12.463	0.000

Student-Newman-Keuls

Live weight	1	3	2
Dry weight	1	3	2
Ash weight	1	3	2
Shell height	1	2	3

*Treatment 1 - crab meal mixed with 25 micron filtered seawater.
Treatment 2 - crab meal mixed with freshwater and 280 grams of rock salt.

these large red areas were present. One such appearance of the red patch occurred directly after the water flow had stopped to all trays during a pump malfunction.

The numbers of heterotrophic and autotrophic flagellates plus diatoms in the tray water receiving crab meal was up to 8 times greater than in the control tray or in the headtank water. Cryptophytes dominated the heterotrophic flagellates in most cases. Bacterial counts of water samples from the tray receiving crab meal varied between 2-4 times the counts of samples from the head tank or the control tray.

Experiment 6

Clams fed crab meal mixed with filtered seawater at 15% clam live weight showed two times the increase in shell height of control clams (Table 15). One-way ANOVA showed that crab meal fed clams had significantly greater final live, dry and ash weights than control clams ($P<0.01$) (Table 16). Crab meal supplemented clams gained twice as much weight as control clams at the completion of 30 days of feeding. Clams receiving crab meal also showed a significantly greater increase in shell height ($P<0.01$) than found in control clams. Supplemented clams showed an increase of 7% of shell height (0.52 mm) at the end of 4 weeks. This growth in shell height is much greater than the growth rates in natural settings for winter at this latitude (Ansell 1968). Mortality was negligible during the experiment although in the nursery during the same time mortality was higher than usual.

Bacterial counts of water from the trays receiving crab meal were 2-3 times greater than those of water samples from the head tank or

TABLE 13. Experiment 5. Initial and final live, dry, and ash weights (mg) and shell heights (mm) of clams held in trays with 156 ml min^{-1} flowing filtered seawater with and without crab meal supplementation for 30 days (n=400 per tray).

	Initial	Final		
		1	2	3
		Crab Meal & SW	Crab Meal & Brine	Control
live weight (mg)				
$\bar{X}$	156.6	176.5	170.1	175.8
SD	6.9	7.9	7.0	7.7
dry weight (mg)				
$\bar{X}$	99.8	114.8	109.2	113.2
SD	4.3	4.4	4.8	5.8
ash weight (mg)				
$\bar{X}$	92.4	106.3	100.8	105.4
SD	4.2	5.0	4.8	4.9
shell height (mm)				
$\bar{X}$	6.9	7.7	7.5	7.2
SD	0.4	0.6	0.5	0.5

TABLE 14. Experiment 5. Comparison of mean final weights and heights of juvenile clams (mean ± 1 SD) held in trays with 156 ml min^{-1} flowing filtered seawater with and without crabmeal supplementation for 30 days (n=400 per tray). F ratios and probabilities are from one-way analysis of variance of final weights and heights. Student-Newman-Keuls (SNK) range tests show subsets of groups at a level of significance $\alpha = 0.05$.

	Treatments				
	1	2	3		
Final	Crab meal (30%)*	Crab meal (30%)*	Control	F ratio	Probability
Live weight (mg)	176.5 (7.9)	170.1 (7.0)	175.8 (7.7)	84.121	0.0000
Dry weight (mg)	114.8 (4.4)	109.2 (4.8)	113.2 (5.8)	125.791	0.0000
Ash weight (mg)	106.3 (5.0)	100.8 (4.8)	105.4 (4.9)	139.736	0.0000
Shell height (mm)	7.7 (0.6)	7.5 (0.5)	7.2 (0.5)	14.883	0.0000

Student-Newman-Keuls

Live weight	2	3	1
Dry weight	2	3	1
Ash weight	2	3	1
Shell height	3	2	1

*Treatment 1 - Crab meal mixed with 10 micron filtered seawater.
Treatment 2 - Crab meal mixed with freshwater and 280 grams of rock salt.

the control tray. Microflagellates made up the majority of the algal species in all samples. Water samples from trays receiving crab meal contained 2-4 times as many microflagellates as was found in samples from the head tank or control tray water.

TABLE 15. Experiment 6. Initial and mean final live, dry, and ash weights (mg) and shell heights (mm) of clams held in trays with 96 ml min^{-1} flowing seawater with and without crab meal supplementation for 30 days (n=400 per tray).

	Initial	Final	
		1	2
		Crab Meal (15%)	Control
live weight (mg)			
$\bar{X}$	158.4	175.2	167.8
SD	8.1	6.4	6.9
dry weight (mg)			
$\bar{X}$	100.7	113.2	106.2
SD	4.4	4.5	5.1
ash weight (mg)			
$\bar{X}$	93.1	104.2	99.2
SD	5.0	4.3	4.2
shell height (mm)			
$\bar{X}$	7.1	7.6	7.1
SD	0.5	0.5	0.6

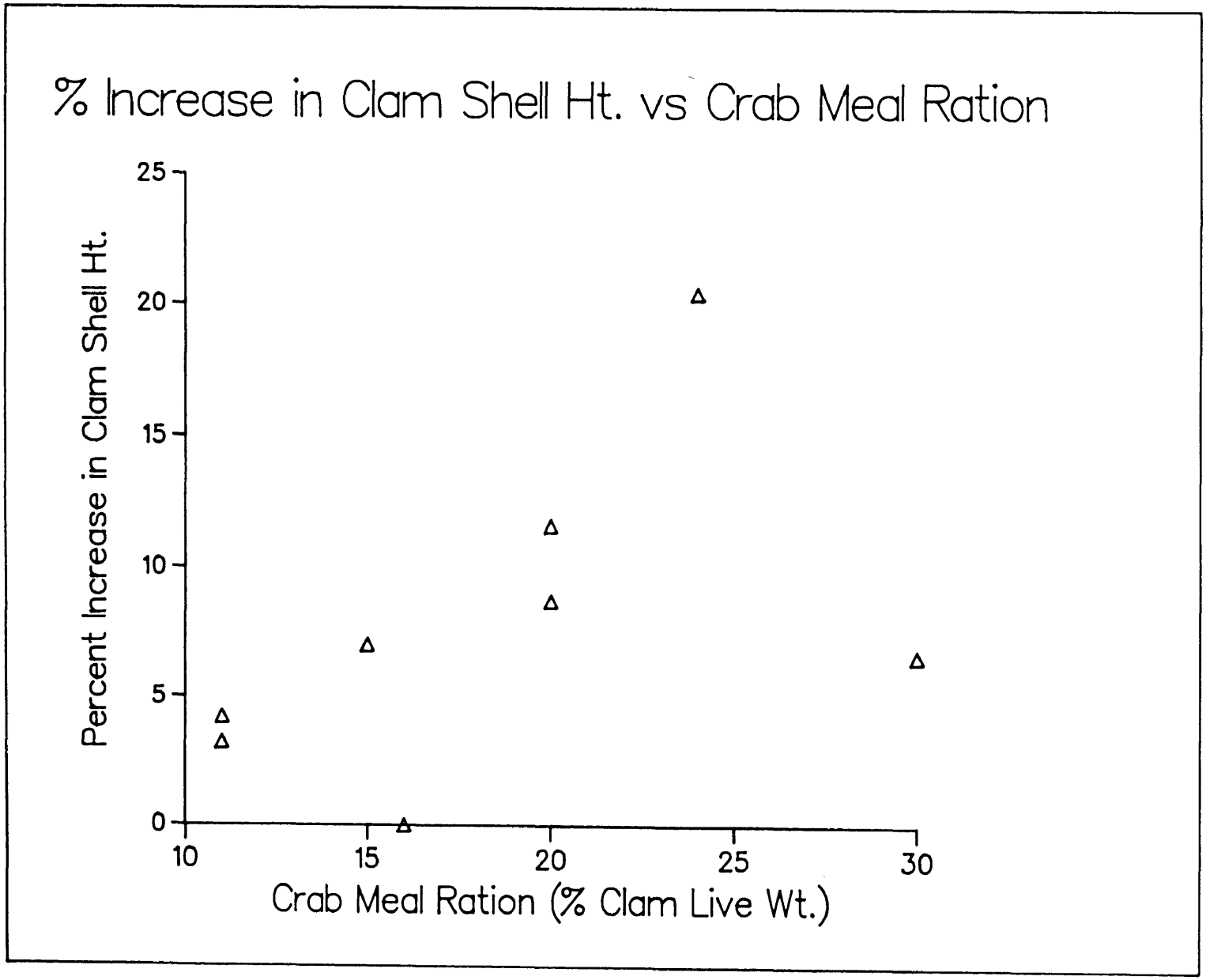
TABLE 16. Experiment 6. Comparison of mean final weights and shell heights of juvenile clams (mean ± 1 SD) held in trays of 96 ml min^{-1} flowing filtered seawater with and without crab meal supplementation (n=400 per tray, duration of experiment = 30 days). F ratios and probabilities are from one-way analysis of variance of final weights and heights.

	Treatments			
	1	2		
Final	Control (0 crab meal)	Crab meal (15% live wt.)	F ratio	Probability
Live weight (mg)	167.8 (6.9)	175.2 (6.4)	246.933	0.0000
Dry weight (mg)	106.2 (5.1)	113.2 (4.5)	432.379	0.0000
Ash weight (mg)	99.2 (4.2)	104.2 (4.3)	267.281	0.0000
Shell height (mm)	7.1 (0.6)	7.6 (0.5)	45.923	0.0000

DISCUSSION

This study indicates the potential for crab meal as a supplemental food for juvenile clams. Increases in shell and tissue growth were found when crab meal was fed at proper rations. Crab meal fed at 11 to 20 percent of clam live weight promoted growth in clams with a shell height of 7 to 9 mm. For 4 to 6 mm size clams, crab meal rations of 18 to 24 percent of clam live weight were fed successfully. Optimum crab meal rations which produced the best growth were 11 to 15 percent of clam live weight for 7-9 mm clams and 20-24 percent for 4-6 mm clams. At the seawater flow rates tested, clams which received greater amounts of crab meal than the optimum rations, showed less growth than clams fed the proper ration. Figure 6 shows the relationship of the increase in clam shell height with crab meal ration. There appeared to be a direct relationship between the increase in shell height and crab meal ration, fed at 11 to 24 percent of clam live weight, in all cases except for TRT 1 in experiment 2. For clams with a shell height of 9.4 mm, crab meal fed at 16 % live weight in TRT 1 was greater than optimum and less growth was seen in these clams than in control clams. When crab meal was fed at 30% of clam live weight less shell growth was observed. This is similiar to the results of decreased growth when greater than optimum amounts of algae are fed to some bivalves. Several authors have determined optimum feeding concentrations of algae for bivalves (Winter 1970, 1976; Tenore and Dunstan 1973). Tenore and Dunstan (1973) found that

Figure 6. The percent increase in shell height of juvenile hard clams at different crab meal rations (ration = percent of clam live weight). Each point represents the mean shell height (mm) of 100 clams.


% Increase in Clam Shell Ht. vs Crab Meal Ration
Percent Increase in Clam Shell Ht.
25
20
15
10
5
0
10
15
20
25
30
Crab Meal Ration (% Clam Live Wt.)

(1973) found that the feeding rate of hard clams increased to a maximum concentration of algae above which the actual feeding rate decreased. They also noted that feeding rates were very depressed at algal concentrations below the optimum level. Similiar responses in feeding behavior occurred in this study. When crab meal concentrations were above the optimum amount, clams with their siphons extended and actively feeding were observed less frequently. Control clams were observed with their shells closed more often than crab meal fed clams (at proper rations) which may have been in response to inadequate amounts of naturally occuring food.

The supplemental feeding of crab meal to juvenile clams resulted in increased growth in weight and shell height regardless of the season or seawater flow rate. Since clams received filtered flowing seawater as in commercial nurseries, natural changes in seawater temperature, food quality, salinity, etc. occurred throughout the year. Chlorophyll *a* levels showed a natural seasonal decrease from summer to winter. The increase in shell height of the control and crab meal fed clams however, appeared inversely related with measured chlorophyll *a* levels (Figure 7). Various seawater flow rates were used in the different experiments. There appeared to be an inverse relationship between the the increase in shell height and the seawater flow rate (Figure 8). Under the usual nursery conditions a direct relationship between seawater flow rate and chlorophyll *a* levels with shell growth would be expected. The data points showing increases in shell height however were largely due to crab meal supplemented clams, since little or no significant shell growth occurred in control clams. Accordingly, it is suspected that both reduced flow rates and

Figure 7. The percent increase in shell height of crab meal supplemented and control clams at different levels of chlorophyll *a* ($\mu g\ l^{-1}$). Each data point represents the mean shell height (mm) of 100 clams.

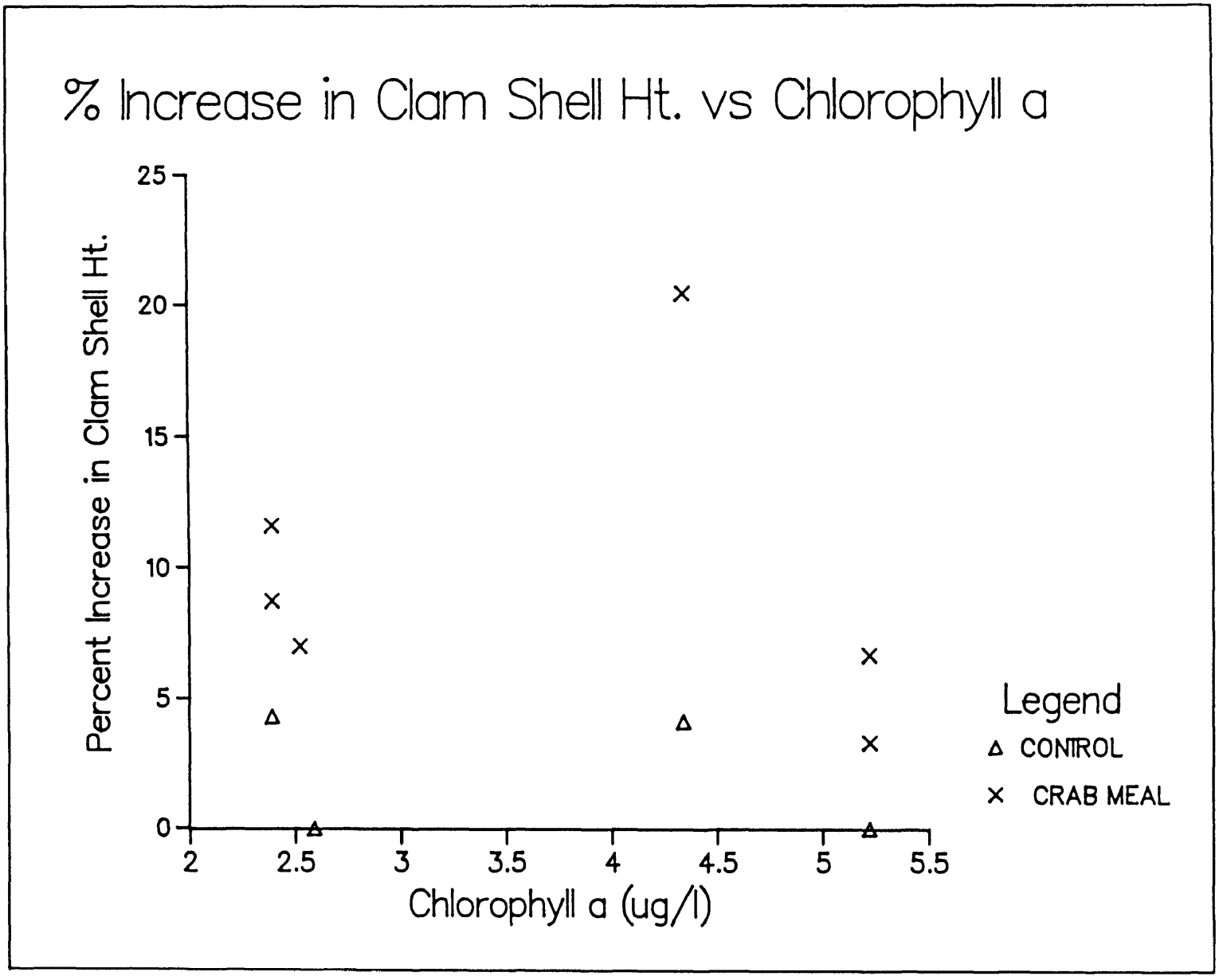
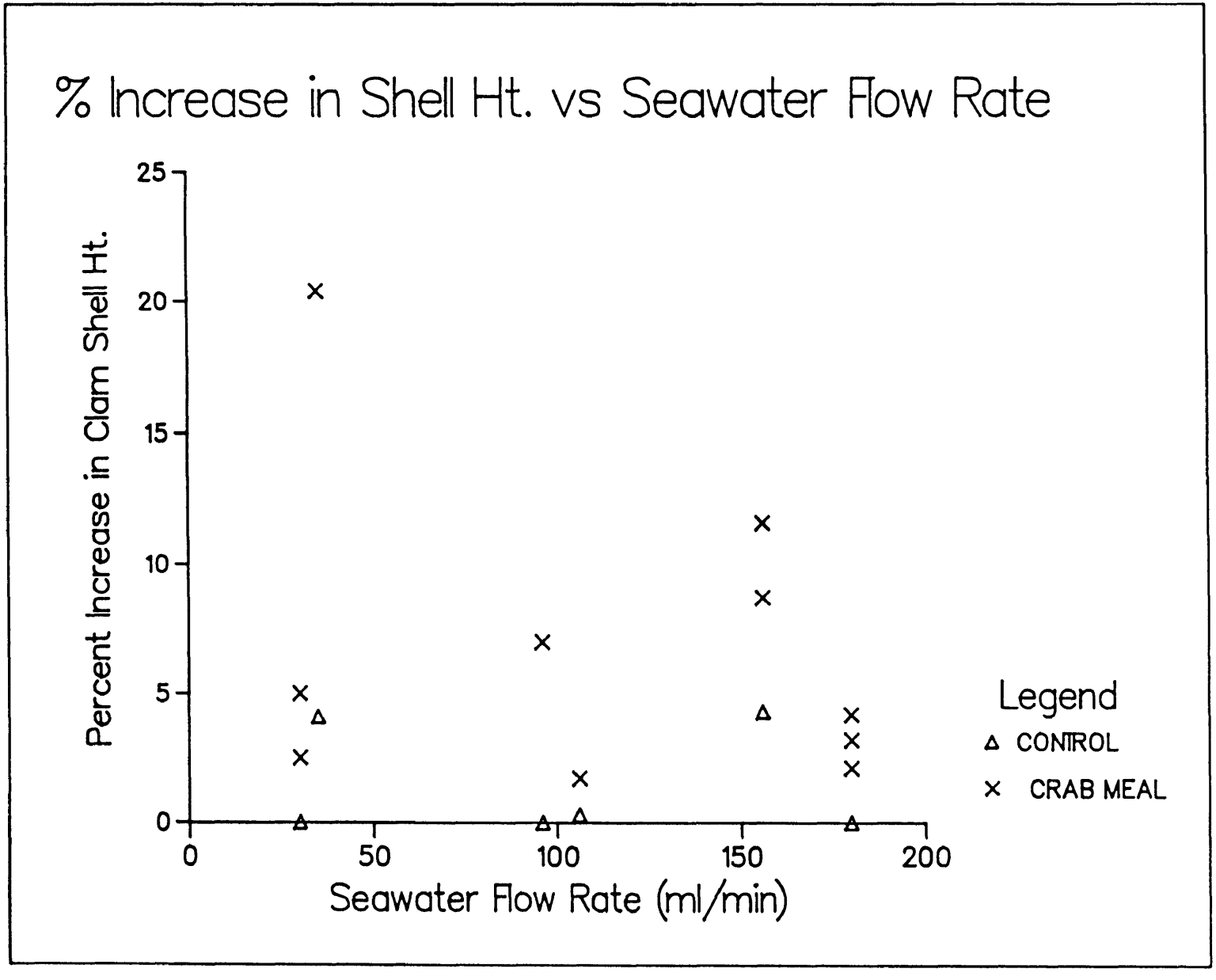

% Increase in Clam Shell Ht. vs Chlorophyll a
Percent Increase in Clam Shell Ht.
25
20
15
10
5
0
2
2.5
3
3.5
4
4.5
5
5.5
Chlorophyll a (ug/l)
Legend
CONTROL
CRAB MEAL

Figure 8. The percent increase in shell height of crab meal supplemented and control clams at different seawater flow rates (ml min^{-1}). Each data point represents the mean shell height (mm) of 100 clams.


% Increase in Shell Ht. vs Seawater Flow Rate
Percent Increase in Clam Shell Ht.
25
20
15
10
5
0
0
50
100
150
200
Seawater Flow Rate (ml/min)
Legend
CONTROL
CRAB MEAL

chlorophyll *a* levels were indicative of a longer residence time of the water in the trays which allowed clams to feed on the crab meal before it was flushed out.

A range of particle sizes of crab meal were fed to juvenile clams in this study. The sieve sizes of 35, 53, 100 and 134 microns were used, each of which formed the upper limit of the particle size range of the crab meal tested. Positive growth results were obtained with all of these sieve sizes in clams fed proper amounts of crab meal. Coulter counter analysis of the 134 micron sieved crab meal showed that 80 per cent of the crab meal was less than 40 microns. Previous authors have noted suitable food particle sizes for bivalves, but have concentrated their effort on the smallest acceptable size (Haven and Morales 1970). Others have shown that larger particle sizes are injested. Foster-Smith (1975) found that Venerupis pullastra exhibited no selection against particles as large as 120 microns. Loosanoff and Engle (1947) found that oysters ingested species of algae up to 60 microns. Ingle et al. (1981) pointed out that the largest size of food accepted by C. virginica or other bivalves has not been determined. In this study all four sizes of crab meal peformed well as a supplemental food.

Various solutions were tested in feeding crab meal. In experiment 4 where the highest crab meal ration fed (30% of clam live weight) was mixed with a brine solution, clams receiving this solution showed greater growth than those receiving crab meal mixed with seawater. Bacterial counts in the trays receiving crab meal were higher in this experiment than in any other examined. The trays receiving crab meal mixed with seawater (TRT 1) had twice the number

of bacteria than in TRT 2 where crab meal was mixed with a brine solution. It is suspected that the brine solution helped keep bacterial numbers from reaching the high numbers observed in TRT 1. Previous workers have noted the detrimental effects of very high bacterial levels on bivalves (Tubiash et al. 1965; Ukeles and Sweeney 1969; Masson 1977). In experiment 5 however, crab meal mixed with seawater produced better clam growth than the same concentration of crab meal mixed with brine. Overall the best clam growth occurred when crab meal was mixed with 25 micron filtered seawater.

A microbial community developed in the food flasks when filtered seawater was mixed with crab meal. A microbial bloom along with crab meal was then delivered to the clams as food. Cryptomonads, small flagellates, and bacteria were observed in higher numbers in the crab meal trays than in the control trays or in the head tank which had only filtered seawater. Cryptomonads and microflagellates are known to be good food organisms for juvenile bivalves (Imai 1949; Iwasaki et al. 1971; Ukeles 1971; Walne 1974; Enright et al. 1986). Some workers have shown that bacteria serve as food for some bivalves (Zobell and Feltham 1938; McHenery et al 1979). It was not possible to determine if the microbial bloom or the crab meal was the most important component of the diet with the experimental design used. Methods of feeding crab meal which keep bacterial and microbial numbers low such as encapsulation of crab meal along with the use of artificial or filtered and sterilized water would be necessary to determine the effects of crab meal as the sole food without the microbial interaction. Antibiotics could also be used to keep bacterial levels down. Additionally the microbial community growing on crab meal could

be fed alone to determine the utilization of these microbes by the clams as food in the absence of crab meal.

The complete nutritional requirements are not known for any bivalve species (Kinne 1977, Langdon 1983a). Philips and Brockway (1956) determined the protein quality of a food by comparing the essential amino acid composition of the food with that of the organism being studied. Table 17 shows the percentage composition of the amino acids of juvenile Mercenaria mercenaria, crab meal, and three algal species that are known to be good food for bivalves. The percentages of amino acids in M. mercenaria are more similiar to crab meal than to the algal species. Crab meal contains higher percentages of the amino acids lysine, histidine, arginine, glutamic acid, cystine, methionine, tyrosine, and phenylalanine than any of the algal species shown. Harrison (1975) has determined the essential amino acids of the mussel, Mytilus californianus. Table 18 presents the essential amino acids of M. californianus along with the percentage composition of those amino acids in M. mercenaria and in crab meal. The essential amino acid composition of the two bivalves is very similiar. Crab meal has comparable or higher levels of these essential amino acids than the juvenile clams except for threonine and lysine. This indicates the good protein quality of crab meal for clams. Other studies have demonstrated requirements of fatty acids for bivalves (Castell and Trider 1980; Langdon and Waldock 1981; Webb and Chu 1983). The consensus of these studies is that $\omega 3$ fatty acids are essential for oysters and the quantity of total $\omega 6$ fatty acids in the diet may affect oyster growth. Crab tissue, which makes up part of the crab meal, has high amounts of $\omega 3$ fatty acids along with the

TABLE 17. The amino acid percentage composition of juvenile Mercenaria mercenaria, crab meal, and three algal species.

Amino Acid	Mercenaria mercenaria (1)	Crab meal	Pyramimonas virginica (2)	Tetraselmis sueica (1)	Isochrysis galbana (3)
*Lysine	7.05	6.29	4.84	5.91	4.95
*Histidine	2.83	2.66	1.58	1.72	1.66
*Arginine	6.00	7.07	4.39	4.31	5.28
Aspartic Acid	11.26	9.78	9.76	9.24	9.49
*Threonine	6.57	4.27	5.81	6.41	3.91
Serine	6.59	4.24	5.81	6.04	5.37
Glutamic Acid	12.09	14.42	12.13	9.61	10.59
*Proline	4.29	7.59	4.88	3.45	5.75
Glycine	7.67	5.56	9.72	1.05	10.62
Alanine	7.52	5.19	11.62	1.31	11.84
Half Cystine	1.79	3.23	0.51	1.17	0.41
*Valine	5.54	4.92	6.41	6.53	7.64
*Methionine	2.53	4.87	0.61	2.59	1.89
*Isoleucine	5.12	4.47	3.98	4.20	5.05
*Leucine	7.19	7.77	8.77	8.63	9.17
Tyrosine	2.60	4.25	2.51	2.22	2.10
*Phenylalanine	3.34	4.56	3.89	4.07	4.08

References: (1) Walne (1970), (2) Webb and Chu (1983), (3) Epifanio (1979).

*Essential amino acid for Mytilus californianus, Harrison (1975).

TABLE 18. The percentage composition of essential amino acids for *Mytilus californianus* and in whole body proteins of *M. californianus*, *Mercenaria mercenaria*, and in crab meal.

	Percent Concentration of Amino Acids		
Amino Acid	*Mytilus californianus* (1)	*Mercenaria mercenaria* (2)	Crab meal
Threonine	7.26	6.57	4.27
Proline	1.67	4.29	7.59
Valine	5.33	5.54	4.92
Methionine	2.03	2.53	4.87
Isoleucine	3.87	5.12	4.47
Leucine	7.42	7.19	7.77
Phenylalanine	2.81	3.34	4.56
Tryptophan	0.22	-	-
Lysine	4.10	7.05	6.30
Histidine	1.03	2.83	2.66
Arginine	4.20	6.00	7.07

References: (1) taken from Harrison (1975), (2) taken from Walne (1970).

presence of ω6 fatty acids (Bonnet et al. 1974). The presence of these fatty acids also indicates the good nutritional quality of crab meal for bivalves.

It is difficult to compare the growth observed in clams supplemented with crab meal to previous experiments with bivalves. For the most part, feeding experiments have been conducted on adult or larval oysters. Oysters have faster growth rates than clams (Claus 1971; Walne 1974) so growth increments occuring during an experiment are not comparable to clam growth. Also the growth rates of juveniles and adult bivalves are very different. Accordingly, it is difficult to compare the growth of adult bivalves fed artificial foods, which have been used in the majority of the cited experiments, to the growth of juvenile clams. Comparison with other feeding studies on juvenile bivalves is also difficult because previous experiments were conducted in beakers or in recirculating water (Murken 1976; Langdon and Bolton 1984; Urban and Langdon 1984). Overall none of the experiments cited earlier have shown that the growth of bivalves fed artifical diets is better than that in natural surroundings or when cultured algae is used. This is the first study to show the successful feeding of an artificial food to juvenile hard clams in a flow-through seawater system.

The results of this study have important implications for the nursery culture of juvenile clams. Clam nurseries are a necessary part of commercial clam culture since clam seed usually cannot be collected in commercial quantities in nature. Clam seed also must be grown to at least 10 mm before planting in field plots to reduce their vulnerability to predators (Castagna and Kraeuter 1981). Food costs

in producing vast quantities of cultured algae or in pumping large volumes of flowing seawater is expensive and is a major factor hindering the commercial viability of clam nurseries. The use of crab meal as a partial or complete replacement for cultured algae could greatly lower food costs. This study showed the effectiveness of crab meal in enhancing clam growth, especially in producing increased shell growth, in conditions similiar to those in commercial nurseries. The diet preparation and feeding methodology employed in this study was very simple and thus easily transferable to commercial operations. Somewhat reduced seawater flow rates were used when feeding crab meal which would represent a savings in pumping costs if applied to commercial operations.

Crab meal was successfully fed during various months of the year with varying amounts of naturally occurring food in the flowing seawater. These results are thus applicable to commercial nurseries where variation in natural food also occurs on a seasonal basis. Increased shell growth occurred in crab meal supplemented clams which is not seen in natural surroundings during late fall or winter. The use of crab meal might allow the overwintering of clam seed too small to be planted in field plots in the fall in nurseries at a reduced cost. Supplemental feeding of crab meal could augment naturally occuring food and would be especially beneficial during periods of time when food levels are seasonally low.

Further studies are required to scale-up and refine the use of crab meal as an artificial food in commercial operations. Experiments need to be conducted to determine whether the microbial bloom which occurs when using crab meal is an important component of the total

food content. Encapsulation of crab meal would allow better testing without the interference of the microbial community. This would prevent leaching problems and reduce the numbers of bacteria occuring in the culture system. Also different methods of delivering crab meal to the culture system such as a dry meal instead of in solution should be tested. Additional experiments are required to determine the optimum crab meal rations for different sizes of clams under different seawater flow rates. Crab meal should also be tested in recirculating seawater systems especially since there were indications that it may have been flushed out before clams were allowed to feed on it in this experiment. The results of this study also indicate that crab meal should be tested as a component in formulated diets. And finally, pilot-scale experiments are required to determine the optimum methods of utilizing crab meal on a commercial scale in a nursery.

SUMMARY AND CONCLUSIONS

The use of natural or cultured algae as a primary food source in commercial clam nurseries is expensive and is one factor preventing the commercial feasability of clam culture. An inexpensive artificial food could reduce costs and allow greater commercial success of the nursery stage of clam culture.

Methods of using crab meal, an inexpensive by-product of the crab-picking industry, as a supplemental food for juvenile hard clams were tested. Experiments were conducted in flow-through seawater systems. Seawater flow rates provided at least enough natural food to sustain clam maintenance activities. Crab meal was mixed with filtered seawater or brine solutions and continuously delivered via a peristaltic pump. Juvenile hard clams with shell heights from 4-9 mm were tested.

The supplemental feeding of crab meal produced positive growth responses in juvenile clams ranging from 4.0 - 9.0 mm (shell height) when fed in proper rations. Clams receiving crab meal showed significantly greater increases in weight and shell height than control clams which fed solely on the natural food contained in the flowing seawater.

Optimum feeding rations of crab meal were determined for different size juvenile clams. Clams with a shell height of 4-6 mm showed the greatest increases in shell height and weight at feeding rates of 20-24% of clam live weight per day. Clams 7-9 mm in height

showed the best growth responses when fed crab meal at 11-15% of clam live weight per day. Crab meal fed at 30% of clam live weight produced less growth in clams than lower rations suggesting that this was an upper limit at the seawater flow rates tested.

Crab meal which was sieved through a 100 or 134 micron mesh, autoclaved, and mixed with 25 micron filtered seawater resulted in the best growth in clams both in terms of weight and shell height.

Increases in shell height and weight did not reflect seasonal changes in water quality or seawater flow rates but was related to crab meal rations and feeding methodology. The amount of crab meal fed was directly correlated with the increase in shell height in clams fed crab meal at rations from 11-24% of clam live weight. With crab meal positive shell growth occurred during months when little or no growth occurs in natural surroundings (November and December). Control clams feeding solely on the natural food contained in the filtered seawater, showed growth more reflective of that observed in natural surroundings for those time periods.

These results are directly applicable to commercial clam nurseries where flow-through seawater systems are utilized. Crab meal, a readily available, inexpensive industrial by-product, could serve as a partial replacement for natural or cultured algae and thus reduce overall food costs. The methods used in this study of feeding crab meal are relatively simple, inexpensive, and readily transferable to a commercial nursery.

Further work is required to refine the use of crab meal as a supplemental food and to test different feeding methodologies. Crab meal should also be tested as a major component in formulated artificial diets.

LITERATURE CITED

Ansell, A. D. 1968. The rate of growth of the hard clam Mercenaria mercenaria (L.) throughout the geographical range. J. Cons. Perm. Int. Explor. Mer. 31:364-409.

Bonnet, J. C., V. D. Sidwell, and E. G. Zook. 1974. Chemical and nutritive values of several fresh and canned finfish, crustaceans, and mollusks. Part II. Fatty Acid Composition. Marine Fish. Rev. 36(2):8-14.

Castagna, M. and J. N. Kraeuter. 1981. Manual for growing the hard clam Mercenaria. Spec. Rep. No. 249, Virginia Institute of Marine Science, Gloucester Point, Virginia.

Castell J. D. and D. J. Trider. 1974. Preliminary feeding trials using artificial diets to study the nutritional requirements of oysters (Crassostrea virginica). J. Fish. Res. Board Can. 31(1):95-99.

Chanley, P. and R. F. Normandin. 1967. Use of artificial foods for larvae of the hard clam, Mercenaria mercenaria (L.) Proc. Nat. Shellfish Ass. 57:31-36.

Chu, F-L. E., K. L. Webb, D. Hepworth, and M. Roberts. 1982. The acceptability and digestibility of microcapsules by larvae of Crassostrea virginica. J. Shellfish Res. 2(1):29-34.

Dunathan, J. P., R. M. Ingle, and W. K. Havens, Jr. 1969. Effects of artificial foods upon oyster fattening with potential commercial applications. Fla. Dep. Nat. Res. Mar. Res. Lab. Tech. Ser. No. 58, 39 p.

Enright, C. T., G. F. Newkirk, J. S. Craigie, and J. D. Castell. 1986. Evaluation of phytoplankton as diets for juvenile Ostrea edulis L. J. Exp. Mar. Biol. Ecol., Vol. 96, pp 1-13.

Epifanio, C. E. 1976. Culture of bivalve molluscs in recirculating systems: Nutritional requirements. Proc. 1st Internat. Conf. Aquaculture Nutrition. Oct. 1975. Lewes, Del. K. S. Price, Jr., W. N. Shaw, and K. S. Danberg (Eds) College of Marine Studies, U. Dela., Newark. 173-180.

_____. 1979. Comparison of yeast and algal diets for bivalves Aquaculture 16:187-192.

Foster-Smith, R. L. 1975. The effect of concentration of suspension on the filtration rates and pseudofaecal production for Mytilus edulis (L.), Cerastoderma edule (L.) and Venerupis pullastra (Montagu). J. Exp. Mar. Biol. Ecol. 17:1-22.

Gabbott, P. A., D. A. Jones, and D. H. Nichols. 1976. Studies on the design and acceptability of microencapsulated diets for marine particle feeders II. Bivalve molluscs p. 127-141 IN: Proc. 10th Eur. Symp. Mar. Biol., Ostend, Beligium Sept 17-23, 1975 Persoone, G. and E. Jaspers (Eds.). Universa Press. Wetteren. Vol. 1 622 p.

Gillespie, L., R. M. Ingle and W. K. Havens, Jr. 1966. Nutritional Studies with adult oysters, Crassostrea virginica (Gmelin). Fla. Bd. Conserv., Tech. Ser. No. 51. 26 pp.

Haas, L. W. 1982. Improved epifluorescence microscopy for observing planktonic micro-organisms. Ann. Inst. Oceanogr. 58(S):261-266.

Haines, K. C. 1973. A rapid technique for recording sizes of juvenile pelecypod molluscs. Aquaculture 1:433.

Harleston, K. R. 1971. The feeding of cornstarch to clams and mussels. M.S. thesis U. of Va., Va. Inst. of Marine Science, Gloucester Pt.

Harrison, C. 1975. The essential amino acids of Mytilus californianus. The Veliger 18:189-193.

Haven, D. S. 1965. Supplemental feeding of oysters with starch. Chesapeake Sci. 6 :43-51.

Haven, D. S. and R. Morales-Alamo. 1970. Filtration of particles from suspension by the American oyster Crassostrea virginica. Biol. Bull. (Woods Hole, Mass.), 139:248-264.

Hayward, D. and K. L. Webb. 1981. Chlorophyll extraction from marine phytoplankton with a dimethyl sulfoxide: acetone: water system. Vir. Inst. of Mar. Sci. Contr. No. 30. 14 pp.

Imai, T. and M. Hatanaka. 1949. On the artificial propagation of the Japanese common oyster Ostrea gigas Thun., by non-colored naked flagellates. Bull. Inst. Agric. Res. Tohoku Univ. 1:33-46.

Ingle, R. M., D. G. Meyer, and M. R. Landrum. 1981. Preliminary notes on a pilot plant for the feeding of adult American oysters. Adelanto Corp. Apalachicola, FL. 137 p.

Iwasaki, H., S. Tanaka and T. Fujiyama. 1971. On Rhodomonas ovalis Nygaard as food for clam larvae. Bull. Jpn. Soc. Sci. Fish., Vol. 37:1044-1048.

Kinne, O. 1977. Marine Ecology vol. III, part 2 Cultivation O. Kinne (Ed.) John Wiley and Sons, New York. 900-35.

Langdon, C. J. 1983a. New techniques and their application to studies of bivalve nutrition. IN: Proc. of the Second International Conference on Aquaculture Nutrition, edited by G. D. Pruder, C. J. Langdon and D. E. Conklin, Louisiana State University, Baton Rouge, Louisiana. pp. 305-320.

Langdon, C. J. 1983b. Growth studies with bacteria-free oyster (Crassostrea gigas) larvae fed on semi-defined artificial diets. Biol. Bull. 164:227-235.

Langdon, C. J. and M. J. Waldock. 1981. The effect of algal and artificial diets on the growth and fatty acid composition of Crassostrea gigas spat. J. Mar. Biol. Ass. U.K. 61:431-448.

_____ and E. T. Bolton. 1984. A microparticulate diet for a suspension-feeding bivalve mollusc, Crassostrea virginica (Gmelin) J. Exp. Mar. Biol. Ecol. 82:239-258.

_____ and C. A. Siegfried. 1984. Progress in the development of artificial diets for bivalve filter feeders. Aquaculture, 39:135-153.

Loosanoff, V. L. and J. B. Engle. 1947. Effect of different concentrations of microorganisms on the feeding of oysters. Fish. Bull. Fish Wildl. Serv. U.S. 51:31-57.

Lubet, P. 1972a. Rapports CNEXO Contrats No. 72/520. Nutrition des Mollusques Bivalves. 12 p.

Lubet, P. 1972b. Rapports CNEXO Contrats No. 72/637. Biologie de la Nutrition et de la Reproduction Chez les Mollusques. 15 p.

MacKenzie, C. L. Jr. 1977. Predation on hard clam (Mercenaria mercenaria) populations. Trans. Am. Fish Soc. 106(6):530-537.

McHenery, J. G., T. H. Birkbeck and J. A. Allen. 1979. The occurrence of lysozyme in marine bivalves. Comp. Biochem. Physiol. 3B:25-28.

McHugh, J. L. 1981. Recent advances in hard clam mariculture. J. of Shellfish Res. (1):51-55.

Masson, M. 1977. Observations sur la nutrition des larves de Mytilus galloprovincialis avec des aliments inerts. Mar. Biol. 40:157-164.

Murken, J. 1976. Feeding experiments with Mytilus edulis L. at small laboratory scale. III Feeding of waste organic products from the fish industry of Bremerhaven as a means of recycling biodegradable wastes. IN: Proceed 10th European symposium on Marine Biology, Ostend, Belgium, 17-23 September 1975. Vol. 1:273-284.

Murray, T. J. and W. D. DuPaul. 1981. Feasibility of crab meal processing in the Chesapeake Bay region. Spec. Rep. in Appl. Mar. Sci. and Ocean. Eng. No. 248. 63 p.

NMFS. 1981. Fisheries of the United States, 1980. Current fishery statistics No. 8100. Prep. by National Marine Fisheries Service, Resource Statistics Div., B. G. Thompson, Chief, Washington, D.C. 132 pp.

Nell, J. A. 1985. Comparison of some single cell proteins in the diet of the Sydney rock oyster (Saccostrea commercialis). Prog. Fish-Cult. 47(2):110-113.

_____ and B. Wisely. 1983. Experimental feeding of Sydney rock oysters (Saccostrea commercialis). II Protein supplementation of artificial diets for adult oysters. Aquaculture 32:1-9.

_____ and B. Wisely. 1984. Experimental feeding of Sydney rock oysters (Saccostrea commercialis). III. Food concentration and fattening procedures. Aquaculture 37:197-208.

Persoone, G. and C. Claus. 1980. Mass culture of algae: a bottleneck in the nursery culturing of molluscs. In: Algae biomass. G. Shelef and G. J. Soeder (Eds.) Elsevier Amsterdam. 265-285.

Philips, A. M. and D. R. Brockway. 1956. The nutrition of trout II. Protein and carbohydrate. The Progressive Fish-Culturist 18:159-164.

Ryther, J. H. 1981. Commercial mariculture in the United States with emphasis on the northeastern region. Inter. Comm. for the Expl. Sea, Mariculture Committee, F:18, 9 p.

Sokal, R. R. and F. J. Rohlf. 1981. Biometry the principles and practise of statistics in Biological Research. W. H. Freeman and Co., San Francisco, 859 p.

Tenore, K. R. and W. M. Dunstan. 1973. Comparison of feeding and biodeposition of three bivalves at different food levels. Mar. Biol. 21:190-195.

Trider, J. T. and D. J. Castell. 1980. Fatty acid requirements of oysters (Crassostrea virginica). J. Nutr. 110:1303-1309.

Tubiash, H., P. Chanley and E. Leifson. 1965. Bacillary necrosis, a disease of larval and juvenile bivalve mollusks. J. Bact. 90:1036-1043.

Turgeon, K. W. and D. S. Haven. 1978. Effects of cornstarch and dextrose on oysters. Veliger 20(4):352-358.

Ukeles, R. 1971. Nutritional requirements in shellfish culture. p. 43-64 In: K. S. Price and D. Maurer (Eds.). Artificial propagation of commercially valuable shellfish. University of Delaware. Newark, Delaware.

Ukeles, R. and B. M. Sweeney. 1969. Influence of dinoflagellate trichocysts and other factors on the feeding of Crassostrea virginica larvae on Monochrysis lutheri. Limnol. Oceanogr. 14:403-410.

Urban, E. R. and C. J. Langdon. 1984. Reduction in costs of diets for the American oyster Crassostrea virginica (Gmelin), by the use of non-algal supplements. Aquaculture 38:277-291.

Wallace, D. E. 1955. The use of equipment and techniques in applied shellfish management. Proc. Nat. Shelf. Assn. 45:209.

Walne, P. R. 1970. Studies on the food value of nineteen genera of algae to juvenile bivalves of the genera Ostrea, Crassostrea, Mercenaria, and Mytilus. Fish. Invest., Lond. Ser II., 26(5) 62 p.

Walne, P. R. 1974. Culture of bivalve molluscs: 50 years experience at Conwy. Fishing News Books Ltd., Farnham, England. 179 p.

Webb, K. L. and F.-L. E. Chu. 1983. Phytoplankton as a food source for bivalve larvae. IN: Proc. of the Second International Conference on Aquaculture Nutrition, edited by G. D. Pruder, C. J. Langdon and D. E. Conklin, Louisiana State University, Baton Rouge, Louisiana. pp. 272-291.

Willis, S. A., W. K. Havens, and R. M. Ingle. 1976. Quality improvement of oysters Crassostrea virginica (Gmelin) using artificial foods. Fla. Dept. Nat. Res., Mar. Res. Lab. Publ. No. 20. 16 pp.

Winter, J. E. 1970. Filter feeding and food utilization in Arctica islandica L. and Modiolus modiolus L. at different food concentrations. p. 196-206 In: Steele, J. H. (Ed.). Marine food chains. Oliver and Boyd, Edinburgh. 552 p.

Winter, J. E. 1974. Growth in Mytilus edulis using different types of food. Ber. dt. wiss. Komm. Meeresforsch. 23:360-375.

Winter, J. E. 1976. Feeding experiments with Mytilus edulis L. at small laboratory scale. II. The influence of suspended silt in addition to algal suspensions on growth. p. 583-600 In: Persoone, G. and E. Jaspers (Eds.). Proc. 10th Eur. Symp. Mar. Biol., Ostend, Belgium, Sept. 17-23, 1975. Universa Press, Welteren. Vol. 1. 620 p.

_____ and R. W. Langton. 1976. Feeding experiments with Mytilus edulis L. at small laboratory scale I. The influence of the total amount of food ingested and food concentration on growth p. 565-581. IN: Pro. Eur. Symp. mar. Biol., Ostend, Belgium Sept 17-23, 1975 Persoone, G. and E. Jaspers (Eds.) Universa Press. Wetteren. Vol 1., 662 p.

Zobell, C. E. and C. G. Feltham. 1938. Bacteria as food for certain marine invertebrates. J. Mar. Res. 312-327.

VITA

PATRICIA LYNN DUNCAN

Born in Christiansburg, Virginia, May 8, 1955. Graduated from Christiansburg High School, Christiansburg, Virginia, June 1973. Attended Virginia Polytechnic Institute and S.U. 1973-1975. Received a B.A. in Biology from the University of South Florida, Tampa, Florida, August, 1977. Entered the School of Marine Science, College of William and Mary as a graduate student in January, 1981.